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Patent and Trademark Office

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9-260

Requestor's Name: Dawn L. Jones Serial Number: 08/541,191

Date: 9/16/96 Phone: 308-4640 Art Unit: 1211

Search Topic:

Please write a detailed statement of search topic. Describe specifically as possible the subject matter to be searched. Define any terms that may have a special meaning. Give examples or relevant citations, authors keywords, etc., if known. For sequences, please attach a copy of the sequence. You may include a copy of the broadest and/or most relevant claim(s).

Please search the following claims:

1, 16, 17, +28

Thanks,

DLJ

STAFF USE ONLY

Date completed: 9-25-96

Searcher: JOHN DANIELMAN

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Number of Searches: 5

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Type of Search

- ____ N.A. Sequence
- ____ A.A. Sequence
- ✓ Structure
- ____ Bibliographic

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- ____ Dialog
- ____ APS
- ____ Geninfo
- ____ SDC
- ____ DARC/Questel
- ____ Other

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=> D HIS

     (FILE 'LREGISTRY' ENTERED AT 06:50:00 ON 25 SEP 96)
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     FILE 'HCAPLUS' ENTERED AT 07:18:47 ON 25 SEP 96
L1            11 S KAYYEM J?/AU
L2            86 S MEADE T?/AU
L3           184 S FRASER S?/AU
L4             3 S L1 AND L2 AND L3
L5           272 S L1 OR L2 OR L3
L6             7 S L5 AND DELIVER?
L7             8 S L4 OR L6
               SELECT L4 1-3 RN

     FILE 'REGISTRY' ENTERED AT 07:20:01 ON 25 SEP 96
L8            16 S E347-362

     FILE 'HCAPLUS' ENTERED AT 07:20:14 ON 25 SEP 96
L9             4 S L7 AND L8
L10            3 S L4 AND L8
L11            5 S L6 NOT L10

=>
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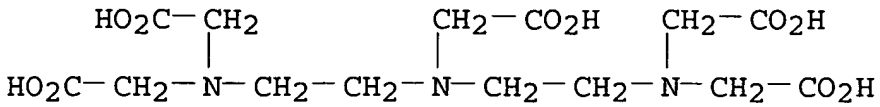
Author

Search

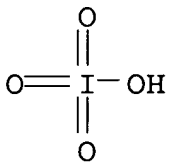
=> D ALL HITSTR L10

L10 ANSWER 1 OF 3 HCAPLUS COPYRIGHT 1996 ACS
AN 1996:404762 HCAPLUS
DN 125:67763
TI Cell-specific gene delivery vehicles for delivery of paramagnetic ions
IN **Kayyem, Jon F.; Meade, Thomas J.; Fraser, Scott E.**
PA California Institute of Technology, USA
SO PCT Int. Appl., 37 pp.
CODEN: PIXXD2
PI WO 9611712 A2 960425
DS W: AL, AM, AT, AU, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IS, JP, KE, KG, KP, KR, KZ, LK, LR, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ
RW: AT, BE, BF, BJ, CF, CG, CH, CI, CM, DE, DK, ES, FR, GA, GB, GR, IE, IT, LU, MC, ML, MR, NE, NL, PT, SE, SN, TD, TG
AI WO 95-US14621 951011
PRAI US 94-321552 941012
DT Patent
LA English
IC ICM A61K047-48
ICS A61K049-00
CC 63-6 (Pharmaceuticals)
Section cross-reference(s): 8
AB A delivery vehicle is described that is capable of being specifically bound to and taken into targeted cells, delivering numerous physiol. agents, particularly paramagnetic ions for magnetic resonance imaging (MRI) of the cells. The delivery vehicle comprises a polymeric mol. having a net pos. charge complexed with another polymeric mol. having a net neg. charge. Cell targeting moieties and physiol. agents, including contrast agents and therapeutic agents, are attached to one or both of the polymeric mols. In one embodiment, the polymeric mol. having a net neg. charge is a nucleic acid. Thus, the delivery vehicles can be used in clin. protocols in which nucleic acids for gene therapy and agents for MRI contrast are co-transported to specific cells allowing medical imaging monitoring of nucleic acid delivery. A suspension of K562 cells were added to a complex of gadolinium-diethylenetriaminepentaacetic acid-polyD-lysine-DNA/transferrin (prepn. given) and allowed to incubate for 10 h at 37.degree.. The controls were simultaneously treated with free transferrin to competitively inhibit the receptor mediated uptake of MRI contrast agent delivery vehicle. MRI images of the cells transfected with particles contg. gadolinium-diethylenetriaminepentaacetic acid-poly-D-lysine showed intense signal indicative of gadolinium contrast enhancement, while the addn. of free transferrin competitively inhibited the uptake of the particles and reduced the MRI contrast.
ST gene delivery vehicle paramagnetic ion; magnetic resonance imaging cell delivery
IT Neoplasm inhibitors
Therapeutics
(cell-specific gene delivery vehicles for delivery of paramagnetic ions)

IT Deoxyribonucleic acids
Transferrins
RL: RCT (Reactant)
(cell-specific gene delivery vehicles for delivery of paramagnetic ions)
IT Polymers, biological studies
RL: THU (Therapeutic use); BIOL (Biological study); USES (Uses)
(cell-specific gene delivery vehicles for delivery of paramagnetic ions)
IT Imaging
(NMR, cell-specific gene delivery vehicles for delivery of paramagnetic ions)
IT Imaging
(contrast agents, cell-specific gene delivery vehicles for delivery of paramagnetic ions)
IT Amines, biological studies
RL: THU (Therapeutic use); BIOL (Biological study); USES (Uses)
(poly-, cell-specific gene delivery vehicles for delivery of paramagnetic ions)
IT **67-43-6**, Diethylenetriaminepentaacetic acid
7790-28-5, Sodium periodate **10138-52-0**, Gadolinium trichloride **25104-18-1**, Poly-L-lysine **25104-18-1D**, Poly-L-lysine, conjugates with transferrins **38000-06-5**, Poly-L-lysine **38000-06-5D**, Poly-L-lysine, conjugates with transferrins
RL: RCT (Reactant)
(cell-specific gene delivery vehicles for delivery of paramagnetic ions)
IT **67-43-6DP**, DTPA, gadolinium and poly-D-lysine complexes
67-43-6DP, Diethylenetriaminepentaacetic acid, reaction products with polylysine **7440-54-2DP**, Gadolinium, DTPA and poly-D-lysine complexes **26853-89-4DP**, Poly-D-lysine, gadolinium and DTPA complexes **26913-90-6DP**, Poly-D-lysine, gadolinium and DTPA complexes
RL: RCT (Reactant); SPN (Synthetic preparation); PREP (Preparation)
(cell-specific gene delivery vehicles for delivery of paramagnetic ions)
IT **124-20-9**, Spermidine **9002-06-6**, Thymidine kinase
60239-18-1, Dota
RL: THU (Therapeutic use); BIOL (Biological study); USES (Uses)
(cell-specific gene delivery vehicles for delivery of paramagnetic ions)
IT **67-43-6**, Diethylenetriaminepentaacetic acid
7790-28-5, Sodium periodate **10138-52-0**, Gadolinium trichloride **25104-18-1**, Poly-L-lysine **25104-18-1D**, Poly-L-lysine, conjugates with transferrins **38000-06-5**, Poly-L-lysine **38000-06-5D**, Poly-L-lysine, conjugates with transferrins
RL: RCT (Reactant)
(cell-specific gene delivery vehicles for delivery of paramagnetic ions)
RN 67-43-6 HCAPLUS
CN Glycine, N,N-bis[2-[bis(carboxymethyl)amino]ethyl]- (7CI, 8CI, 9CI) (CA INDEX NAME)

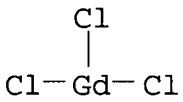


RN 7790-28-5 HCAPLUS
CN Periodic acid (HIO4), sodium salt (8CI, 9CI) (CA INDEX NAME)



● Na

RN 10138-52-0 HCAPLUS
CN Gadolinium chloride (GdCl3) (6CI, 7CI, 8CI, 9CI) (CA INDEX NAME)

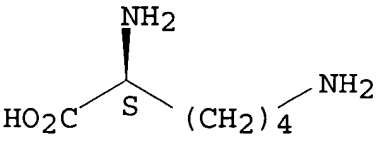


RN 25104-18-1 HCAPLUS
CN L-Lysine, homopolymer (9CI) (CA INDEX NAME)

CM 1

CRN 56-87-1
CMF C6 H14 N2 O2
CDES 5:L

Absolute stereochemistry.

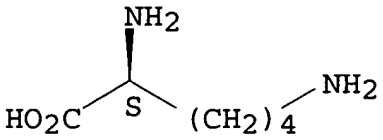


RN 25104-18-1 HCAPLUS
CN L-Lysine, homopolymer (9CI) (CA INDEX NAME)

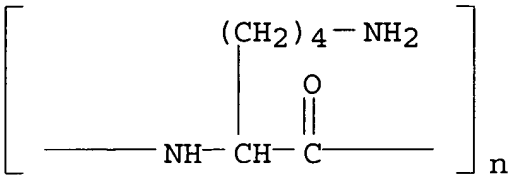
CM 1

CRN 56-87-1
CMF C6 H14 N2 O2
CDES 5:L

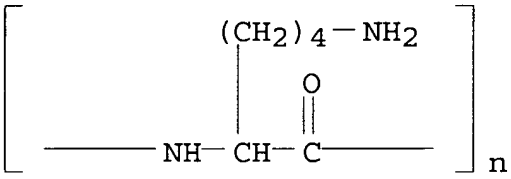
Absolute stereochemistry.



RN 38000-06-5 HCAPLUS
CN Poly[imino[1-(4-aminobutyl)-2-oxo-1,2-ethanediyl]], (S)- (9CI) (CA INDEX NAME)

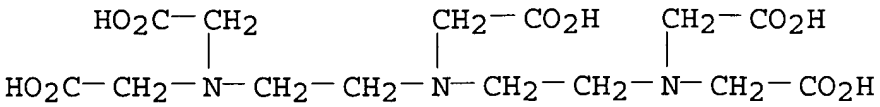


RN 38000-06-5 HCAPLUS
CN Poly[imino[1-(4-aminobutyl)-2-oxo-1,2-ethanediyl]], (S)- (9CI) (CA INDEX NAME)



IT **67-43-6DP**, DTPA, gadolinium and poly-D-lysine complexes
7440-54-2DP, Gadolinium, DTPA and poly-D-lysine complexes
26853-89-4DP, Poly-D-lysine, gadolinium and DTPA complexes
26913-90-6DP, Poly-D-lysine, gadolinium and DTPA complexes
RL: RCT (Reactant); SPN (Synthetic preparation); PREP (Preparation)
(cell-specific gene delivery vehicles for delivery of paramagnetic ions)

RN 67-43-6 HCAPLUS
CN Glycine, N,N-bis[2-[bis(carboxymethyl)amino]ethyl]- (7CI, 8CI, 9CI) (CA INDEX NAME)



RN 7440-54-2 HCAPLUS
CN Gadolinium (8CI, 9CI) (CA INDEX NAME)

Gd

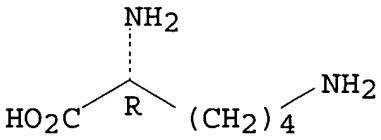
RN 26853-89-4 HCAPLUS

CN D-Lysine, homopolymer (9CI) (CA INDEX NAME)

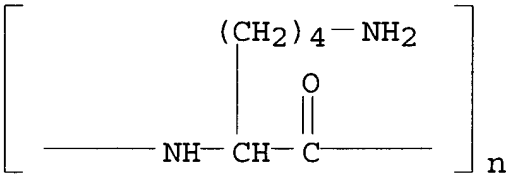
CM 1

CRN 923-27-3
CMF C6 H14 N2 O2
CDES 5:D

Absolute stereochemistry.



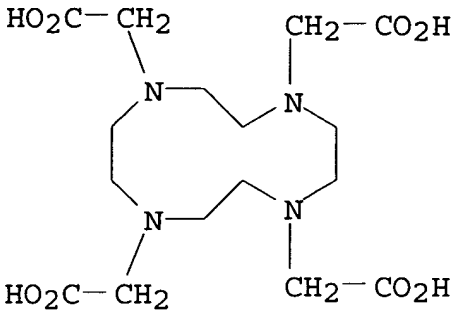
RN 26913-90-6 HCAPLUS
CN Poly[imino[1-(4-aminobutyl)-2-oxo-1,2-ethanediyl]], (R)- (9CI) (CA INDEX NAME)



IT **124-20-9**, Spermidine **9002-06-6**, Thymidine kinase **60239-18-1**, Dota
RL: THU (Therapeutic use); BIOL (Biological study); USES (Uses)
(cell-specific gene delivery vehicles for delivery of paramagnetic ions)
RN 124-20-9 HCAPLUS
CN 1,4-Butanediamine, N-(3-aminopropyl)- (8CI, 9CI) (CA INDEX NAME)

RN 9002-06-6 HCAPLUS
CN Kinase (phosphorylating), thymidine (9CI) (CA INDEX NAME)

*** STRUCTURE DIAGRAM IS NOT AVAILABLE ***
RN 60239-18-1 HCAPLUS
CN 1,4,7,10-Tetraazacyclododecane-1,4,7,10-tetraacetic acid (9CI) (CA INDEX NAME)


HO2C— CH2
CH2— CO2H
N
N
N
N
HO2C— CH2
CH2— CO2H

=> D ALL HITSTR L10 2

L10 ANSWER 2 OF 3 HCAPLUS COPYRIGHT 1996 ACS
AN 1995:931249 HCAPLUS
DN 123:334352
TI Nucleic acid mediated electron transfer
IN **Meade, Thomas J.; Kayyem, Jon F.; Fraser, Scott E.**
PA California Institute of Technology, USA
SO PCT Int. Appl., 58 pp.
CODEN: PIXXD2
PI WO 9515971 A2 950615
DS W: AM, AT, AU, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, JP, KE, KG, KP, KR, KZ, LK, LR, LT, LU, LV, MD, MG, MN, MW, NL, NO, NZ, PL, PT, RO, RU, SD, SE, SI, SK, TJ, TT, UA, UZ, VN
RW: AT, BE, BF, BJ, CF, CG, CH, CI, CM, DE, DK, ES, FR, GA, GB, GR, IE, IT, LU, MC, ML, MR, NE, NL, PT, SE, SN, TD, TG
AI WO 94-US13893 941205
PRAI US 93-166036 931210
DT Patent
LA English
IC ICM C07H021-00
ICS G01N033-50; C12Q001-68
CC 9-15 (Biochemical Methods)
Section cross-reference(s): 76
AB The present invention provides for the selective covalent modification of nucleic acids with redox active moieties such as transition metal complexes. Electron donor and electron acceptor moieties are covalently bound to the ribose-phosphate backbone of a nucleic acid at predetd. positions. The resulting complexes represent a series of new derivs. that are bimol. templates capable of transferring electrons over very large distances at extremely fast rates. These complexes possess unique structural features which enable the use of an entirely new class of bioconductors and photoactive probes. Prepn. of 5'-2'-ruthenium bisbipyridineimidazole-aminouridine-GCTACGA was demonstrated. A method for the synthesis of long DNA duplexes with electron transfer moieties at the 5'-termini was also described.
ST bioconductor photoactive probe nucleic acid; electron transfer DNA duplex
IT Electric conductors
(bioconductor; nucleic acid mediated electron transfer and its application in bioconductors and photoactive probes)
IT Deoxyribonucleic acids
Nucleic acids
RL: BUU (Biological use, unclassified); NUU (Nonbiological use, unclassified); SPN (Synthetic preparation); BIOL (Biological study); PREP (Preparation); USES (Uses)
(conjugates of single-stranded nucleic acid with redox active moieties; nucleic acid mediated electron transfer and its application in bioconductors and photoactive probes)
IT Transition metals, biological studies
RL: BUU (Biological use, unclassified); NUU (Nonbiological use, unclassified); BIOL (Biological study); USES (Uses)
(nucleic acid mediated electron transfer and its application in bioconductors and photoactive probes)

IT Nucleotides, biological studies
RL: BUU (Biological use, unclassified); NUU (Nonbiological use, unclassified); SPN (Synthetic preparation); BIOL (Biological study); PREP (Preparation); USES (Uses)
(oligo-, photoactive probes; nucleic acid mediated electron transfer and its application in bioconductors and photoactive probes)

IT **170572-27-7P 170572-28-8P**
RL: NUU (Nonbiological use, unclassified); SPN (Synthetic preparation); PREP (Preparation); USES (Uses)
(prepn. of; DNA capable of mediating electron transfer and its application in bioconductors and photoactive probes)

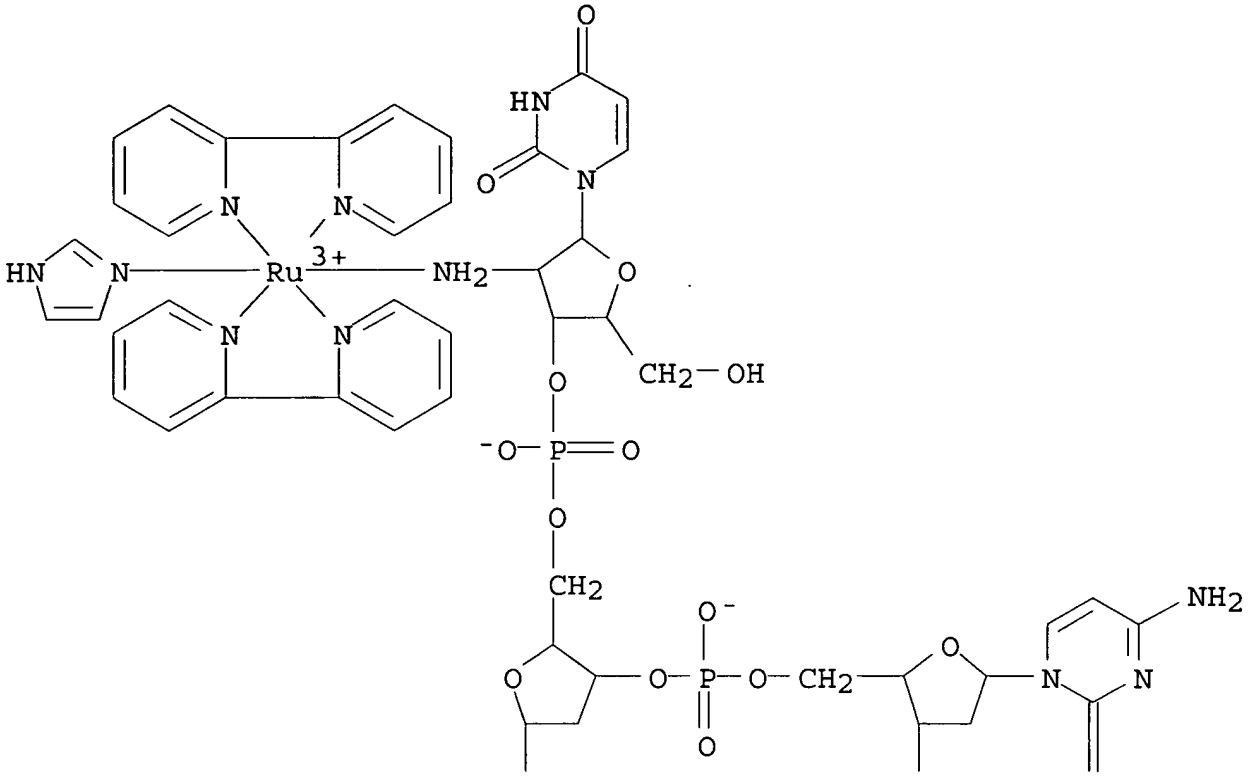
IT **135896-91-2P 170572-25-5P 170572-26-6P**
RL: NUU (Nonbiological use, unclassified); SPN (Synthetic preparation); PREP (Preparation); USES (Uses)
(prepn. of; for prepn. of DNA capable of mediating electron transfer and its application in bioconductors and photoactive probes)

IT **170572-27-7P 170572-28-8P**
RL: NUU (Nonbiological use, unclassified); SPN (Synthetic preparation); PREP (Preparation); USES (Uses)
(prepn. of; DNA capable of mediating electron transfer and its application in bioconductors and photoactive probes)

RN 170572-27-7 HCAPLUS

CN Ruthenate(4-), [2'-amino-2'-deoxyuridylyl-(3'.fwdarw.5')-2'-deoxyguanylyl-(3'.fwdarw.5')-2'-deoxycytidylyl-(3'.fwdarw.5')-thymidylyl-(3'.fwdarw.5')-2'-deoxyadenylyl-(3'.fwdarw.5')-2'-deoxycytidylyl-(3'.fwdarw.5')-2'-deoxyguanylyl-(3'.fwdarw.5')-2'-deoxyadenosinato(7-)]bis(2,2'-bipyridine-N,N')(1H-imidazole-N3)-, heptahydrogen (9CI) (CA INDEX NAME)

PAGE 1-A



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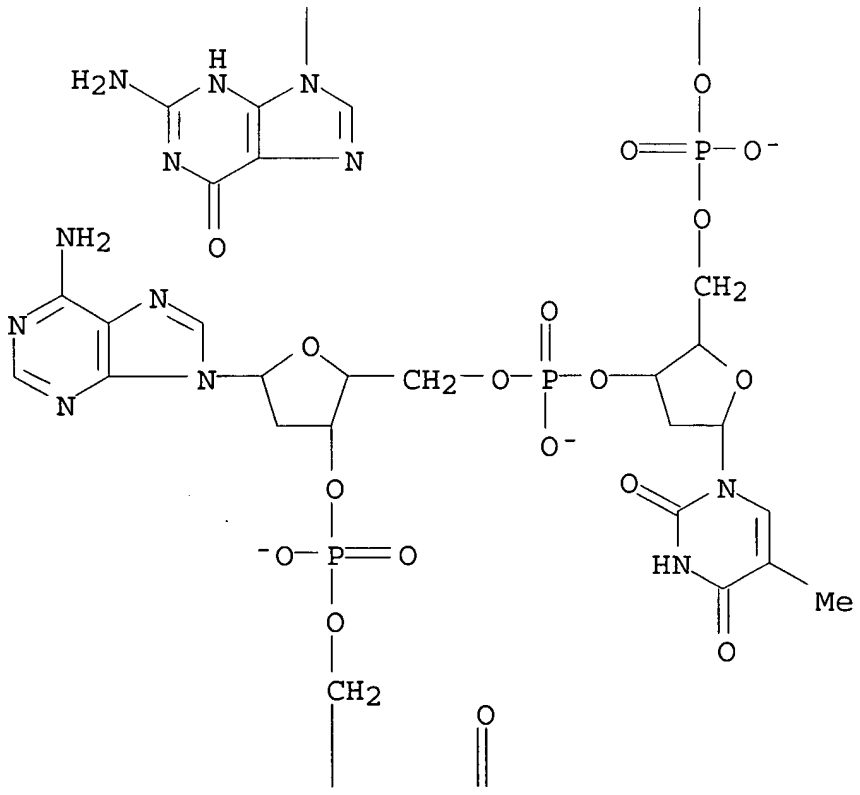

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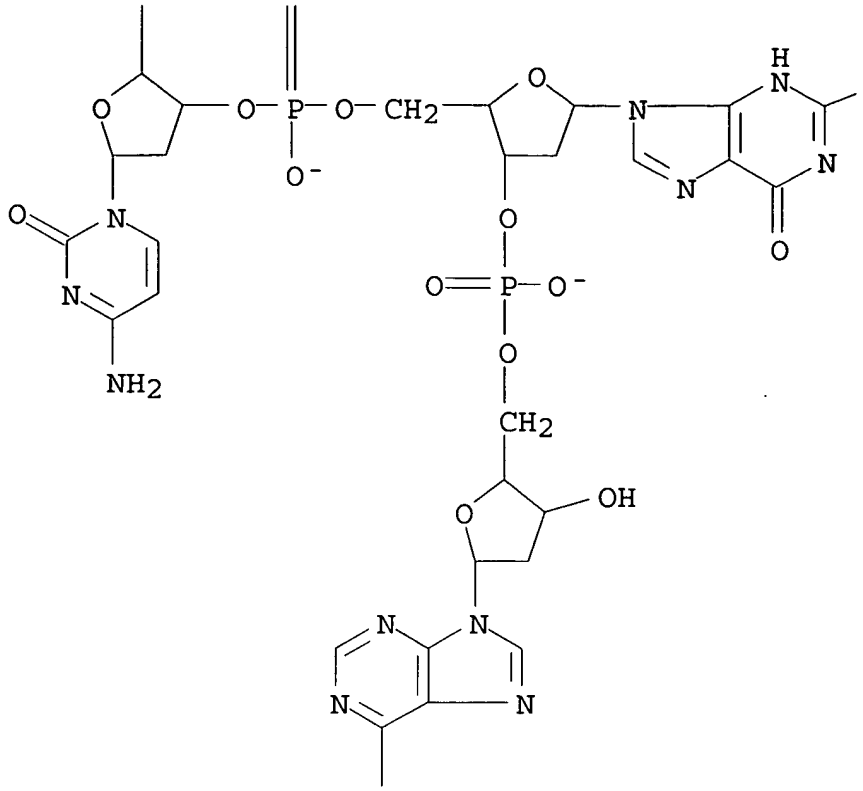

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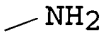
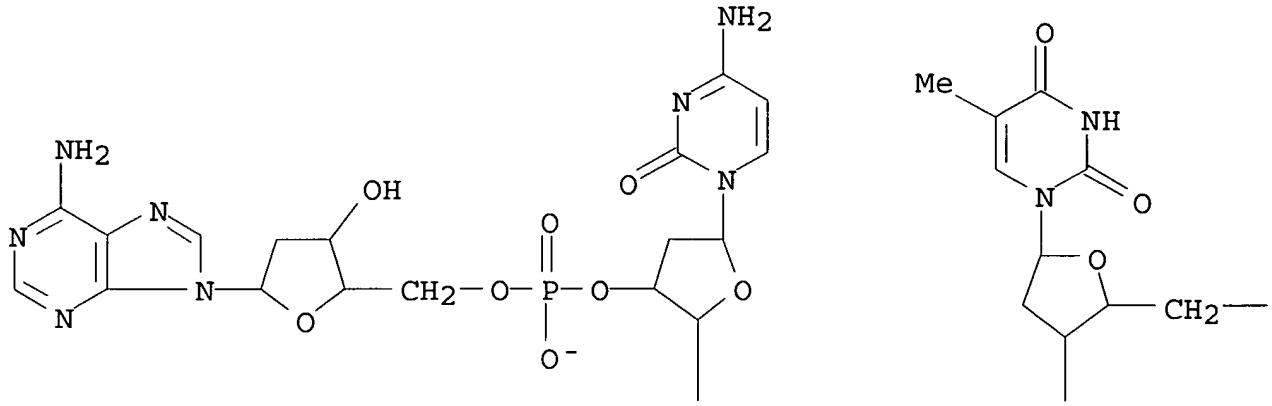


PAGE 4-A

● 7 H^+

RN 170572-28-8 HCAPLUS
CN Ruthenate(4-), [2'-amino-2'-deoxyuridylyl-(3'.fwdarw.5')-2'-deoxycytidylyl-(3'.fwdarw.5')-2'-deoxyguanylyl-(3'.fwdarw.5')-thymidylyl-(3'.fwdarw.5')-2'-deoxyadenylyl-(3'.fwdarw.5')-2'-deoxyguanylyl-(3'.fwdarw.5')-2'-deoxycytidylyl-(3'.fwdarw.5')-2'-deoxyadenosinato(7-)]tetraammine(pyridine)-, heptahydrogen (9CI) (CA INDEX NAME)

PAGE 1-A



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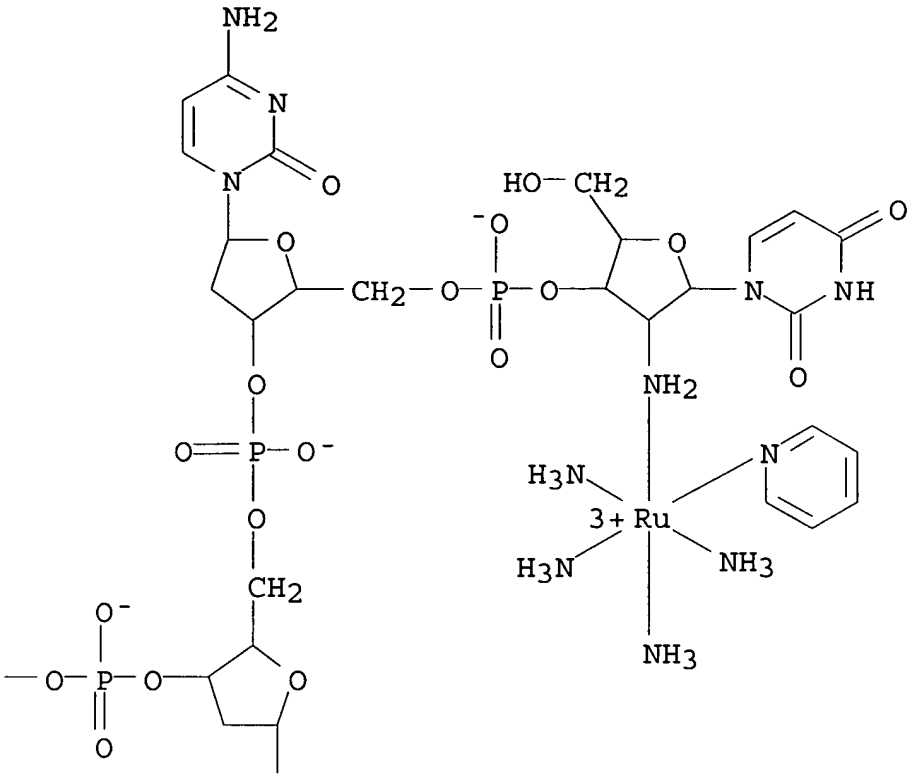

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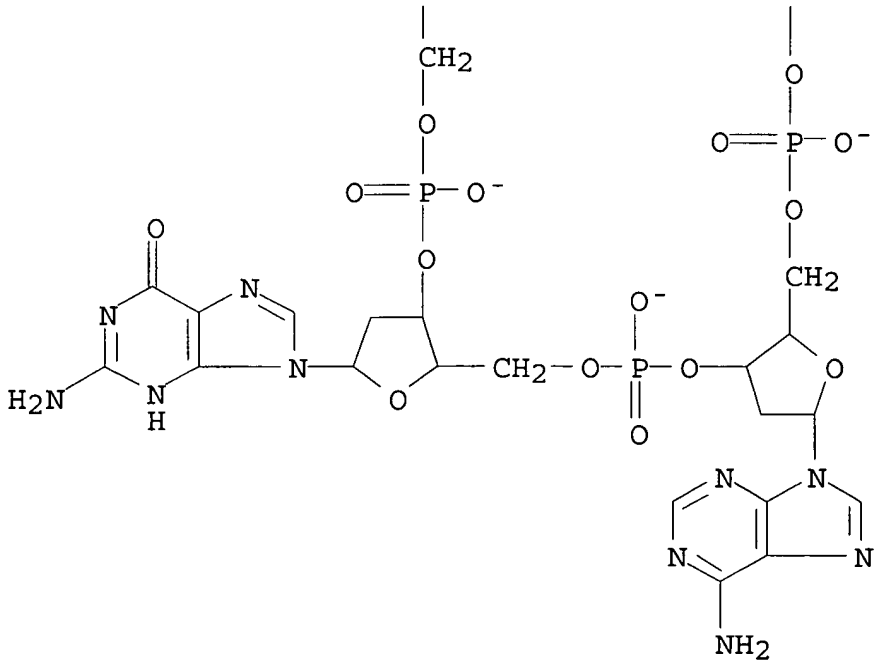

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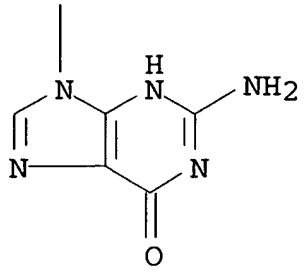
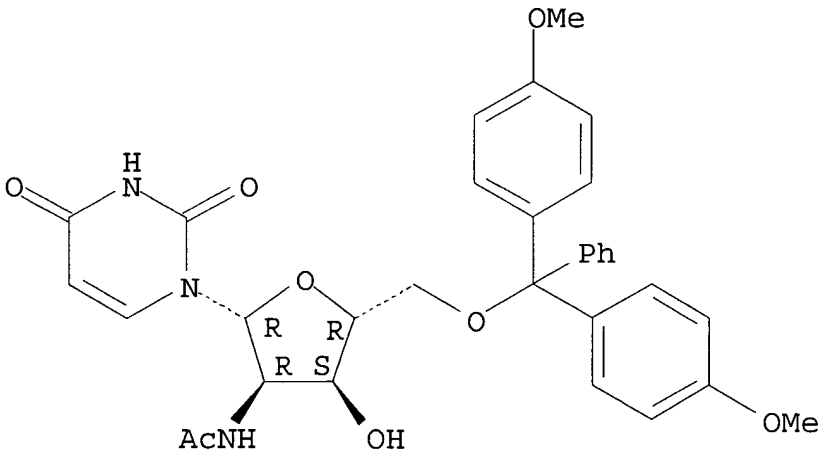


● 7 H^+

IT **135896-91-2P 170572-25-5P 170572-26-6P**
RL: NUU (Nonbiological use, unclassified); SPN (Synthetic preparation); PREP (Preparation); USES (Uses)
(prepn. of; for prepn. of DNA capable of mediating electron transfer and its application in bioconductors and photoactive probes)

RN 135896-91-2 HCAPLUS
CN Uridine, 2'-(acetylamino)-5'-O-[bis(4-methoxyphenyl)phenylmethyl]-2'-deoxy- (9CI) (CA INDEX NAME)

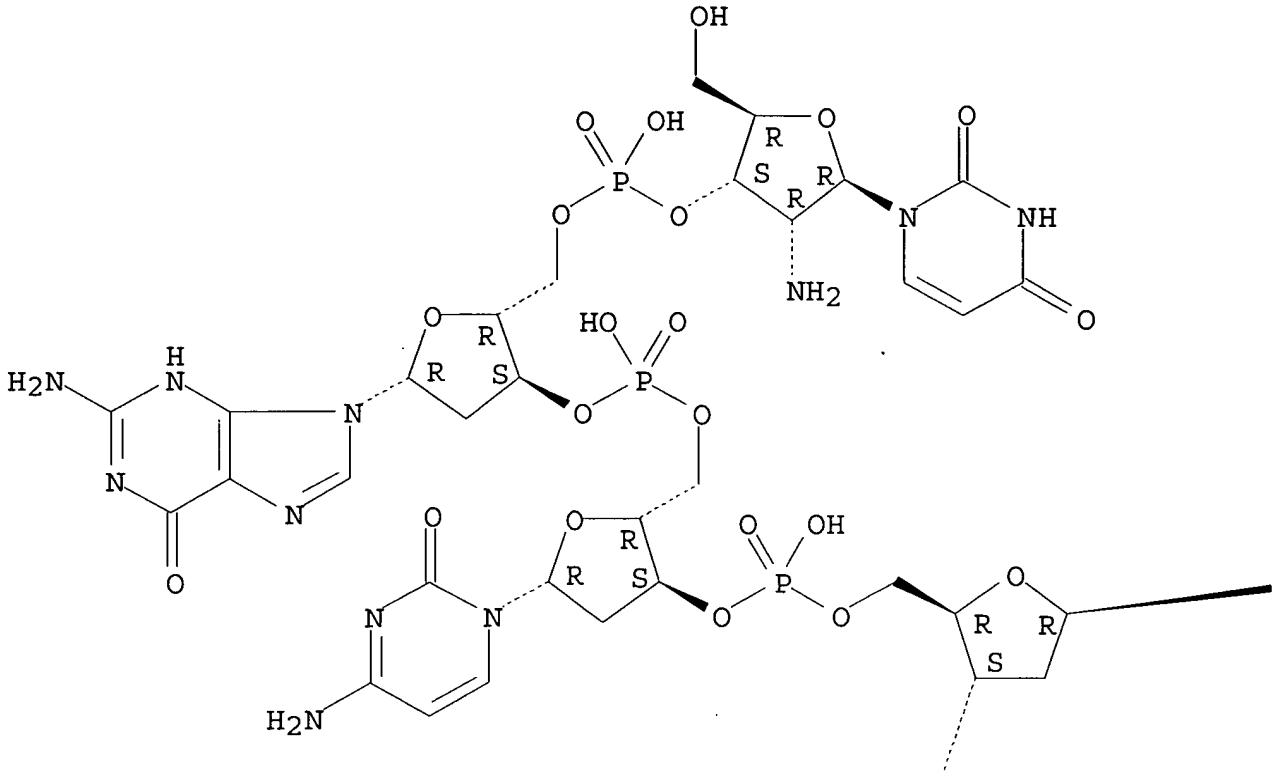
Absolute stereochemistry.



RN 170572-25-5 HCAPLUS
CN Adenosine, 2'-amino-2'-deoxyuridylyl-(3'.fwdarw.5')-2'-deoxyguanylyl-(3'.fwdarw.5')-2'-deoxycytidylyl-(3'.fwdarw.5')-thymidylyl-(3'.fwdarw.5')-2'-deoxyadenylyl-(3'.fwdarw.5')-2'-deoxycytidylyl-(3'.fwdarw.5')-2'-deoxyguanylyl-(3'.fwdarw.5')-2'-deoxy- (9CI) (CA INDEX NAME)

Absolute stereochemistry.

PAGE 1-A



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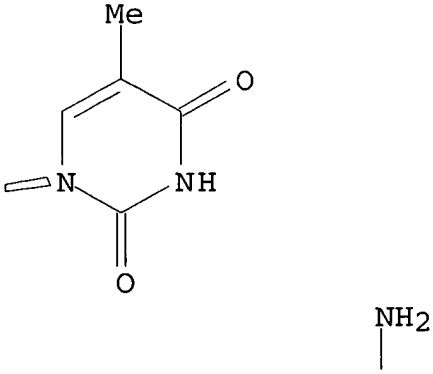

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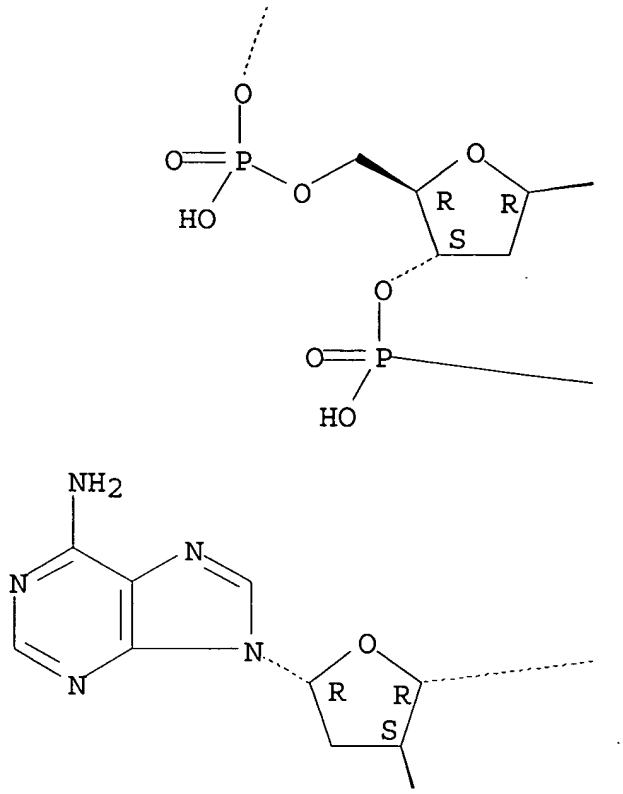
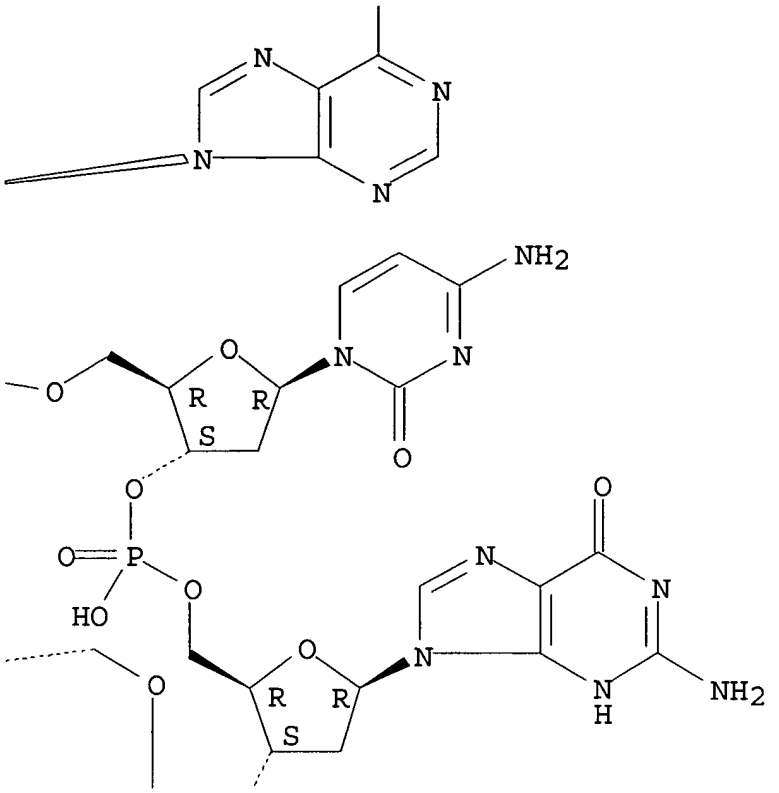
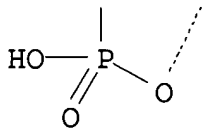


PAGE 2-B



PAGE 3-A

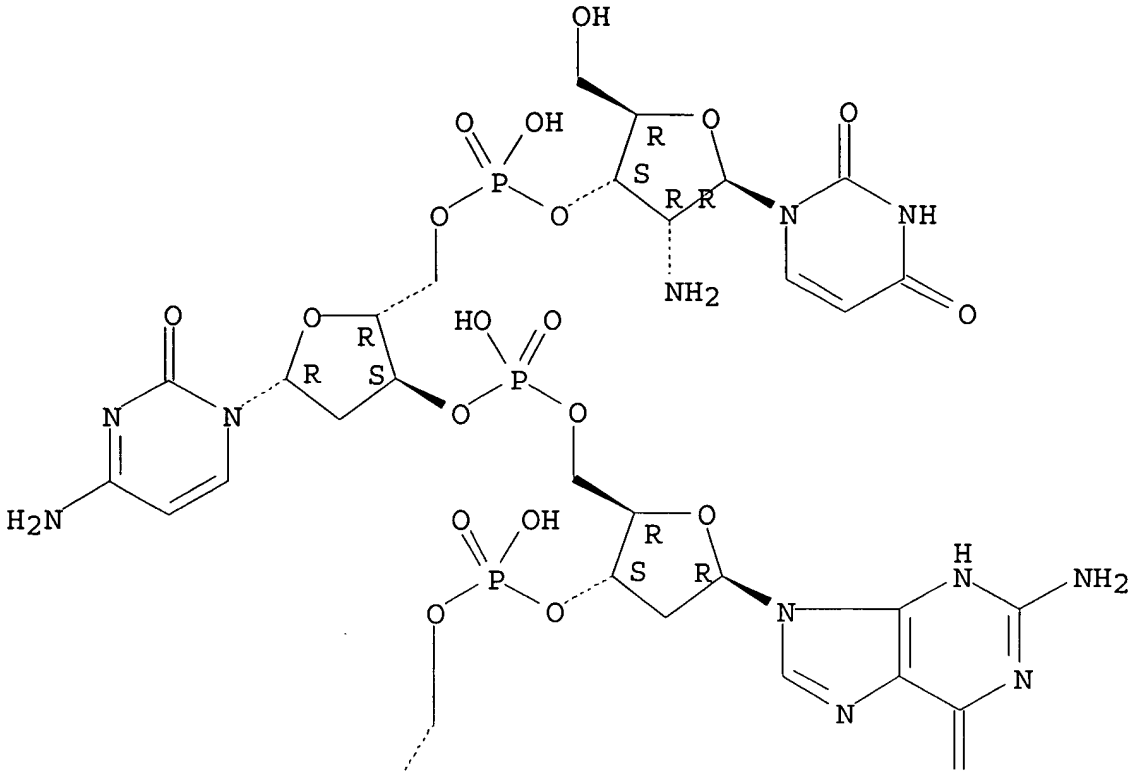
PAGE 3-B



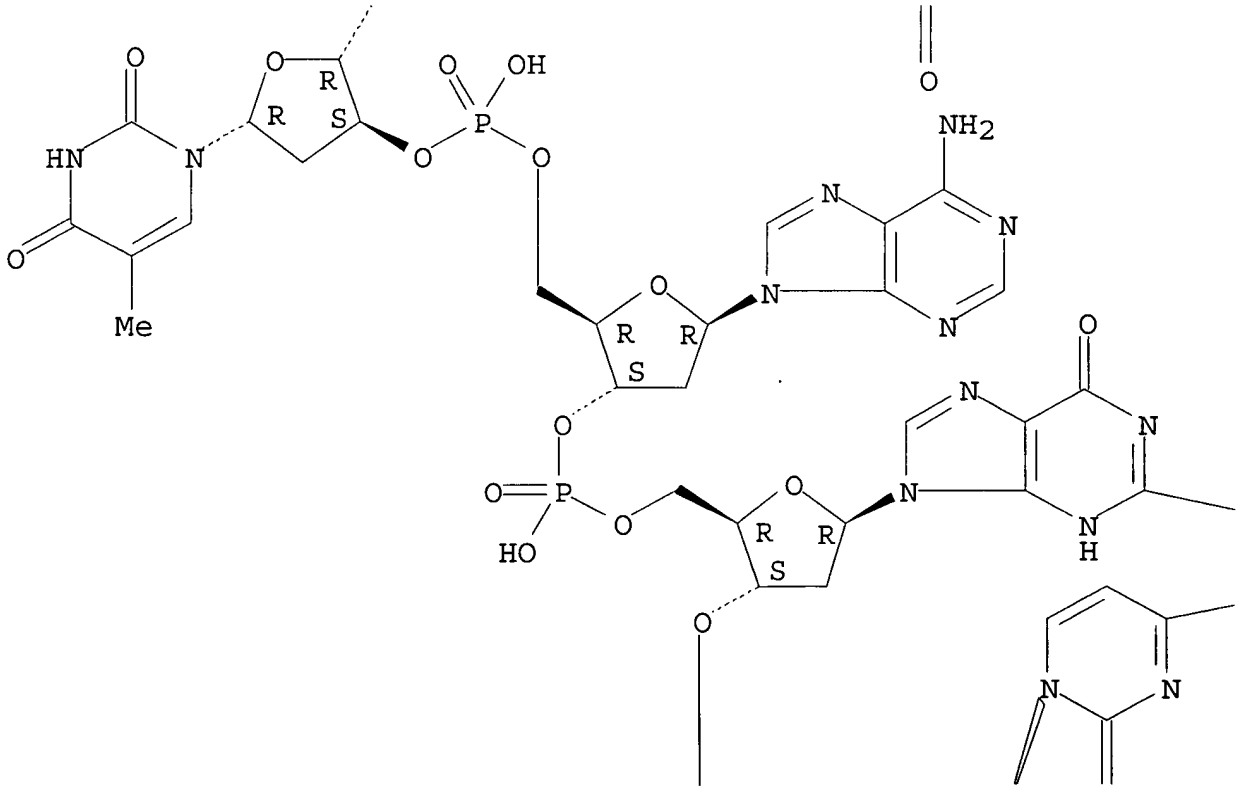
RN 170572-26-6 HCAPLUS
CN Adenosine, 2'-amino-2'-deoxyuridylyl-(3'.fwdarw.5')-2'-deoxycytidylyl-(3'.fwdarw.5')-2'-deoxyguanylyl-(3'.fwdarw.5')-thymidylyl-(3'.fwdarw.5')-2'-deoxyadenylyl-(3'.fwdarw.5')-2'-deoxyguanylyl-(3'.fwdarw.5')-2'-deoxycytidylyl-(3'.fwdarw.5')-2'-deoxy- (9CI) (CA INDEX NAME)

Absolute stereochemistry.

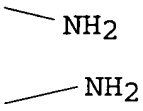
PAGE 1-A



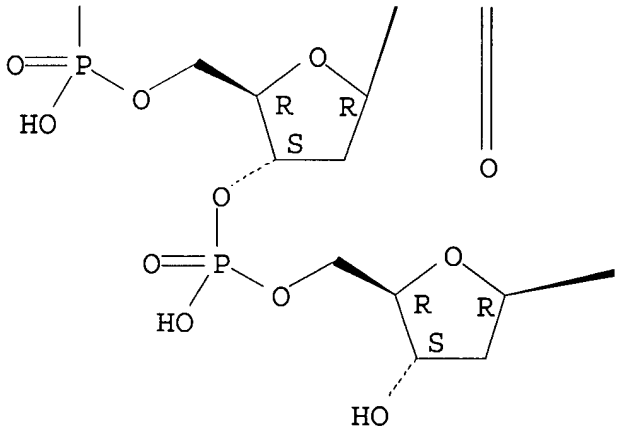
PAGE 2-A



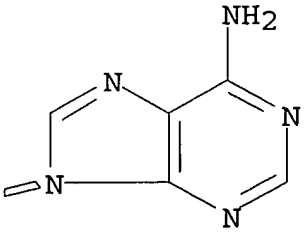
PAGE 2-B



PAGE 3-A



PAGE 3-B



=> D ALL HITSTR L10 3

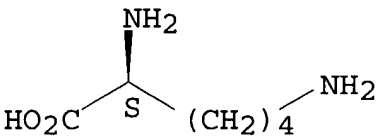
L10 ANSWER 3 OF 3 HCAPLUS COPYRIGHT 1996 ACS
AN 1995:861079 HCAPLUS
DN 123:279808
TI Receptor-targeted co-transport of DNA and magnetic resonance contrast agents
AU **Kayyem, Jon Faiz**; Kumar, Roshan M.; **Fraser, Scott E.**; **Meade, Thomas J.**
CS Div. Biology, Beckman Inst., California Inst. Technology, Pasadena, CA, 91125, USA
SO Chem. Biol. (1995), 2(9), 615-20
CODEN: CBOLE2; ISSN: 1074-5521
DT Journal
LA English
CC 8-1 (Radiation Biochemistry)
AB Ligand mols. conjugated to polylysine can be electrostatically bound to DNA and can bind receptors or antigens on the surface of cells, delivering the DNA into specific cells and tissues. Several researchers have used this approach to generate non-viral vehicles for the efficient delivery of DNA to specific cells. We have attempted to adopt this general approach to the cell-specific delivery of magnetic contrast agents for use in magnetic resonance imaging (MRI). We have synthesized a new class of agents capable of both transfecting genes into cells and enhancing the contrast of the targeted cells for MRI. DNA is used both to encode a marker gene and as a mol. scaffold, which electrostatically binds polylysine conjugated to transferrin, an iron uptake protein, and polylysine modified with gadolinium chelated to diethylenetriamineepetaacetic acid. When cells displaying the transferrin receptor are treated with these particles, high levels of gene expression are obsd., higher than with control particles composed only of transferrin, polylysine and DNA. The treated cells show specific MRI contrast enhancement, which did not require expression of the marker gene. The development of this class of particles permits the use of novel protocols by which genes for genetic therapy and agents for MRI contrast are co-transported. These protocols may allow non-invasive MRI monitoring of DNA delivery for gene therapy in real time.
ST DNA magnetic resonance contrast agent; gene magnetic resonance imaging polylysine transferrin
IT Biological transport
Nuclear magnetic resonance
(receptor-targeted co-transport of DNA and magnetic resonance contrast agents)
IT Transferrins
RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
(receptor-targeted co-transport of DNA and magnetic resonance contrast agents)
IT Deoxyribonucleic acids
RL: BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(receptor-targeted co-transport of DNA and magnetic resonance contrast agents)
IT Imaging
(contrast agents, receptor-targeted co-transport of DNA and magnetic resonance contrast agents)

IT Therapeutics
(geno-, receptor-targeted co-transport of DNA and magnetic resonance contrast agents)
IT **25104-18-1**, Polylysine
RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
(receptor-targeted co-transport of DNA and magnetic resonance contrast agents)
IT **25104-18-1**, Polylysine
RL: BPR (Biological process); BSU (Biological study, unclassified); BIOL (Biological study); PROC (Process)
(receptor-targeted co-transport of DNA and magnetic resonance contrast agents)
RN 25104-18-1 HCAPLUS
CN L-Lysine, homopolymer (9CI) (CA INDEX NAME)

CM 1

CRN 56-87-1
CMF C6 H14 N2 O2
CDES 5:L

Absolute stereochemistry.



=> D ALL 1-5

L11 ANSWER 1 OF 5 HCAPLUS COPYRIGHT 1996 ACS
AN 1995:982344 HCAPLUS
DN 124:11399
TI Storage-stable composition containing fabric softener and perfume-carrier mixture
IN **Fraser, Stuart Bernard**; Parsons, John Stuart; Willis, Edwin
PA Unilever PLC, UK; Unilever N. V.
SO PCT Int. Appl., 21 pp.
CODEN: PIXXD2
PI WO 9522594 A1 950824
DS W: AM, AT, AU, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, JP, KE, KG, KP, KR, KZ, LK, LR, LT, LU, LV, MD, MG, MN, MW, MX, NL, NO, NZ, PL, PT, RO, RU, SD, SE, SI, SK, TJ, TT, UA, UG
RW: AT, BE, BF, BJ, CF, CG, CH, CI, CM, DE, DK, ES, FR, GA, GB, GR, IE, IT, LU, MC, ML, MR, NE, NL, PT, SE, SN, TD, TG
AI WO 95-EP624 950217
PRAI GB 94-3242 940221
DT Patent
LA English
IC ICM C11D003-50
ICS C11D001-62
CC 46-5 (Surface Active Agents and Detergents)
OS MARPAT 124:11399
AB The title compn. with good perfume stability is prepd. by forming an aq. dispersion of a cationic softening agent and adding a mixt. of a perfume and a carrier substance (esp. tallow oil or palm oil) with slip point <45.degree.. The compn. provides good **delivery** of perfume to fabrics.
ST glyceride carrier perfume softener fabric; tallow oil carrier perfume softener fabric; palm oil carrier perfume softener fabric; storage stability perfume softener fabric
IT Palm oil
RL: TEM (Technical or engineered material use); USES (Uses)
(perfume carrier in storage-stable compns. contg. cation fabric softeners)
IT Fats and Glyceridic oils
RL: TEM (Technical or engineered material use); USES (Uses)
(perfume carriers in storage-stable compns. contg. cation fabric softeners)
IT Perfumes
Softening agents
(storage-stable compns. contg. perfume-glyceridic carrier mixts. and cationic fabric softeners)
IT Quaternary ammonium compounds, uses
RL: TEM (Technical or engineered material use); USES (Uses)
(ester group-contg., fabric softeners; storage-stable compns. contg. perfume-carrier mixts. and)
IT Tallow
RL: TEM (Technical or engineered material use); USES (Uses)
(oil, perfume carrier in storage-stable compns. contg. cation fabric softeners)
IT 34004-36-9D, (2,3-Dihydroxypropyl)trimethylammonium chloride, esters with hydrogenated tallow fatty acids

RL: TEM (Technical or engineered material use); USES (Uses)
(fabric softeners; storage-stable compns. contg. perfume-carrier mixts. and)

L11 ANSWER 2 OF 5 HCAPLUS COPYRIGHT 1996 ACS
AN 1995:790861 HCAPLUS
DN 123:219574
TI Transfection of Folate-Polylysine DNA Complexes: Evidence for Lysosomal **Delivery**
AU Mislick, Kimberly A.; Baldeschwieler, John D.; **Kayyem, Jon F.**; **Meade, Thomas J.**
CS Division of Chemistry and Chemical Engineering, California Institute of Technology, Pasadena, CA, 91125, USA
SO Bioconjugate Chem. (1995), 6(5), 512-15
CODEN: BCCHES; ISSN: 1043-1802
DT Journal
LA English
CC 3-2 (Biochemical Genetics)
Section cross-reference(s): 13
OS CJACS
AB We are utilizing the folate receptor for the intracellular **delivery** of DNA. In this study, a folate-poly-L-lysine (FPLL) conjugate was synthesized and equilibrated with plasmid DNA encoding the firefly luciferase gene. The FPLL-DNA complexes were added to KB cells treated with chloroquine. Luciferase activity of cells incubated with FPLL-DNA was 6-fold higher than of cells exposed to poly-L-lysine (PLL)-DNA. The addn. of free folic acid competitively inhibited the enhancement of gene expression. Removal of chloroquine from the media significantly inhibited transfection efficiency of FPLL-DNA complexes. We conclude that FPLL-DNA complexes are **delivered** into KB cells via folate receptor-mediated endocytosis and likely follow a lysosomal pathway into the cytoplasm.
ST transfection folate polylysine DNA receptor lysosome
IT Gene, animal
RL: BPR (Biological process); BIOL (Biological study); PROC (Process)
(luciferase; folate-poly-L-lysine (FPLL) conjugate was synthesized and equilibrated with plasmid DNA encoding the firefly luciferase gene for lysosomal **delivery**)
IT Lysosome
Transformation, genetic
(transfection of folate-polylysine DNA complexes: evidence for lysosomal **delivery**)
IT Deoxyribonucleic acids
RL: BPR (Biological process); BUU (Biological use, unclassified); BIOL (Biological study); PROC (Process); USES (Uses)
(transfection of folate-polylysine DNA complexes: evidence for lysosomal **delivery**)
IT Animal cell line
(KB, folate-poly-L-lysine-DNA complexes are **delivered** into KB cells via folate receptor-mediated endocytosis and likely follow a lysosomal pathway into the cytoplasm)
IT Receptors
RL: BAC (Biological activity or effector, except adverse); BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(folic acid, folate-poly-L-lysine-DNA complexes are **delivered** into KB cells via folate receptor-mediated

endocytosis and likely follow a lysosomal pathway into the cytoplasm)

IT 9014-00-0, Luciferase
RL: BSU (Biological study, unclassified); BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(folate-poly-L-lysine (FPLL) conjugate was synthesized and equilibrated with plasmid DNA encoding the firefly luciferase gene for lysosomal **delivery**)

IT 54-05-7, Chloroquine
RL: BAC (Biological activity or effector, except adverse); BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(folate-poly-L-lysine-DNA complexes were added to KB cells treated with chloroquine for receptor-mediated endocytotic transfection)

IT 59-30-3, biological studies 25104-18-1, Polylysine
RL: BAC (Biological activity or effector, except adverse); BUU (Biological use, unclassified); BIOL (Biological study); USES (Uses)
(transfection of folate-polylysine DNA complexes: evidence for lysosomal **delivery**)

L11 ANSWER 3 OF 5 HCAPLUS COPYRIGHT 1996 ACS
AN 1994:426407 HCAPLUS
DN 121:26407
TI Modulation of doxorubicin efficacy in P388 leukemia following co-administration of verapamil in mini-osmotic pumps
AU Slate, Doris L.; **Fraser-Smith, Elizabeth B.**; Rosete, Jose D.; Freitas, Vicki R.; Kim, Yong N.; Casey, Sharon M.
CS Syntex Discovery Res., Inst. Biochem. and Cell Biol., Palo Alto, CA, 94304, USA
SO In Vivo (1993), 7(6A), 519-23
CODEN: IVIVE4; ISSN: 0258-851X
DT Journal
LA English
CC 1-6 (Pharmacology)
Section cross-reference(s): 63
AB Co-administration of doxorubicin and verapamil in Alzet mini-osmotic pumps increased the survival of B6D2F1 mice bearing the multidrug-resistant P388/ADR leukemia. A range of doxorubicin and verapamil combinations was studied to define dose-dependent efficacy and toxicity. High doses of doxorubicin (10 mg/kg/day) and verapamil (150 mg/kg/day) could be administered alone without any effect on survival. However, combining high doses of these two agents resulted in host toxicity. Doxorubicin doses of 1-10 mg/kg/day in combination with verapamil at 25-100 mg/kg/day were found to improve survival, compared with either agent alone. Combination therapy also improved the survival of mice bearing the drug-sensitive P388/0 leukemia, when compared to anthracycline treatment alone. The efficacy of the min-osmotic pump **delivery** protocol was compared with other regimens **delivering** the same total cumulative dose of doxorubicin via repeated i.p. injections.
ST miniosmotic pump verapamil doxorubicin leukemia inhibition; multidrug resistant leukemia doxorubicin verapamil
IT Neoplasm inhibitors
(leukemia, doxorubicin-verapamil combination, mini-osmotic pump in relation to)
IT Drug resistance
(multi-, of P388/ADR leukemia, treatment of, with

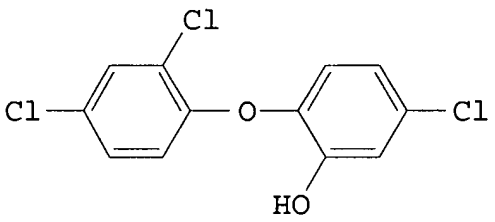
doxorubicin-verapamil combination, mini-osmotic pump in relation to)

IT Pharmaceutical dosage forms
(osmotic pumps, miniaturized, in doxorubicin-verapamil combination treatment of multidrug-resistant and drug-sensitive P388 leukemia)

IT 52-53-9, Verapamil
RL: BIOL (Biological study)
(doxorubicin and, in treatment of multidrug-resistant and drug-sensitive P388 leukemia, mini-osmotic pump in relation to)

IT 23214-92-8, Doxorubicin
RL: BIOL (Biological study)
(verapamil and, in treatment of multidrug-resistant and drug-sensitive P388 leukemia, mini-osmotic pump in relation to)

L11 ANSWER 4 OF 5 HCAPLUS COPYRIGHT 1996 ACS
AN 1987:38214 HCAPLUS
DN 106:38214
TI Oral disposition of triclosan (2,4,4'-trichloro-2'-hydroxydiphenyl ether) **delivered** from a dentifrice
AU Gilbert, R. J.; **Fraser, S. B.**; Van der Ouderaa, F. J. G.
CS Unil. Res. Port Sunlight Lab., Gibbs Dent. Div., Bebington/Wirral/Merseyside, UK
SO Caries Res. (1987), 21(1), 29-36
CODEN: CAREBK; ISSN: 0008-6568
DT Journal
LA English
CC 62-7 (Essential Oils and Cosmetics)
Section cross-reference(s): 1, 63
GI



I

AB The oral retention and intra-oral distribution of triclosan (I) [3380-34-5] in man were detd. after use of an antiplaque toothpaste slurry contg. this antibacterial. I was detd. by HPLC. Thirty-eight percent of the I in a 1 g dose of toothpaste (0.50%, wt./wt. I) and 62% of a 3 g dose was retained. I was detected and measured in plaque and this antibacterial was also present in saliva. Levels of approx. 20-30 .mu.g I/mL of saliva were measured 5 min after toothpaste use and these had fallen to approx. 3-4 .mu.g/mL 2 h later. A significant inhibition of plaque acid prodn. followed use of the antiplaque dentifrice.

ST dentifrice triclosan oral disposition

IT Drug bioavailability
(of triclosan, from dentifrices in humans, acid formation in relation to)

IT Dentifrices
(triclosan **delivered** from, oral disposition of, in

humans)
IT Mouth
(triclosan disposition in, from dentifrice in humans)
IT Saliva
(triclosan uptake by, from dentifrice in humans)
IT 3380-34-5, Triclosan
RL: PROC (Process)
(oral disposition of, from dentifrice in humans)

L11 ANSWER 5 OF 5 HCAPLUS COPYRIGHT 1996 ACS
AN 1977:579267 HCAPLUS
DN 87:179267
TI The influence of ammonia on the oxygen **delivery** system of coho salmon hemoglobin
AU Sousa, Robert J.; **Meade, Thomas L.**
CS Dep. Anim. Sci., Univ. Rhode Island, Kingston, R. I., USA
SO Comp. Biochem. Physiol. A (1977), 58(1A), 23-8
CODEN: CBPAB5
DT Journal
LA English
CC 6-3 (General Biochemistry)
Section cross-reference(s): 12
AB Spectrophotometric data showed that prolonged exposure of coho salmon (Oncorhynchus kisutch) to high levels of NH3 had a neg. effect on the O-carrying capacity of Hb. The toxic effect was induced by the accumulation of acid metabolites in the blood after an enzymic stimulation of glycolysis by NH4+ and a simultaneous suppression of the tricarboxylic acid cycle.
ST ammonia Hb Oncorhynchus
IT Oncorhynchus kisutch
(Hb of, oxygen affinity of, ammonia effect on)
IT Glycolysis
Tricarboxylic acid cycle
(ammonia effect on, in coho salmon, Hb in relation to)
IT Methemoglobins
RL: BIOL (Biological study)
(in coho salmon, ammonia effect on)
IT Hemoglobins
RL: PRP (Properties)
(oxygen affinity of, ammonia effect on, in coho salmon)
IT 7782-44-7, biological studies
RL: PRP (Properties)
(Hb affinity of, ammonia effect on, in coho salmon)
IT 7664-41-7, biological studies
RL: BIOL (Biological study)
(Hb oxygen affinity response to, in coho salmon)

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=>    D HIS

     (FILE 'HCAPLUS' ENTERED AT 07:20:14 ON 25 SEP 96)
                DEL HIS   Y
L1          48268 S DELIVER?
L2            622 S L1 AND (POS OR POSITIV? OR POLYCATION? OR CATION?) AND
L3             17 S L2 AND (POLYMAINE# OR SPERMIDINE? OR POLYLYSINE OR POLY
L4             52 S L2 AND (POLYNUCLEO? OR DNA OR NUCLEIC OR OLIGONUCLEO?)
L5              5 S L2 AND POLYAMINE?
L6             19 S L3 OR L5
L7              9 S L6 AND L4
L8             21 S L2 AND CELL####(3A)TARGET?
L9              6 S L8 AND POLYMER?
L10            13 S L7 OR L9
L11             7 S L8 AND (L3 OR L4 OR L5)
L12            13 S L7 OR L9 OR L11
L13             7 S L4 AND CELL####(3A)TARGET?
L14             1 S L13 AND (CONTRAST? OR IMAG?)
L15           855 S  L1 AND CELL####(3A)TARGET?
L16           263 S RECEPTOR? AND L15
L17            12 S L16 AND L2
L18             8 S L2 AND CELL####(4A)UPTAKE?
L19             0 S L18 AND HYDROPHOB?
L20             0 S L14 NOT L12
L21             0 S L13 NOT L12
L22             8 S L17 NOT L12
L23             6 S L18 NOT (L12 OR L17)
                  SAV JONES/L ALL

     FILE 'WPIDS' ENTERED AT 08:24:28 ON 25 SEP 96
L24             8 S L12
L25             1 S L14
L26             1 S L17
L27             6 S L18
L28            10 S L24-L27

     FILE 'MEDLINE' ENTERED AT 08:31:41 ON 25 SEP 96
L29             9 S L12
L30             0 S L14
L31             9 S L17
L32             6 S L18
L33            21 S L29-L32

     FILE 'BIOSIS' ENTERED AT 08:37:37 ON 25 SEP 96
L34            11 S L12
L35             0 S L14
L36            10 S L17
L37             7 S L18
L38            23 S L34-L37

     FILE 'HCAPLUS' ENTERED AT 08:42:45 ON 25 SEP 96
L39            27 S L12 OR L14 OR L17 OR L18

     FILE 'BIOSIS' ENTERED AT 08:42:53 ON 25 SEP 96
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Text search

```
=> D HIS

     (FILE 'HCAPLUS' ENTERED AT 07:20:14 ON 25 SEP 96)
                DEL HIS   Y
L1          48268 S DELIVER?
L2            622 S L1 AND (POS OR POSITIV? OR POLYCATION? OR CATION?) AND
L3             17 S L2 AND (POLYMAINE# OR SPERMIDINE? OR POLYLYSINE OR POLY
L4             52 S L2 AND (POLYNUCLEO? OR DNA OR NUCLEIC OR OLIGONUCLEO?)
L5              5 S L2 AND POLYAMINE?
L6             19 S L3 OR L5
L7              9 S L6 AND L4
L8             21 S L2 AND CELL####(3A)TARGET?
L9              6 S L8 AND POLYMER?
L10            13 S L7 OR L9
L11             7 S L8 AND (L3 OR L4 OR L5)
L12            13 S L7 OR L9 OR L11
L13             7 S L4 AND CELL####(3A)TARGET?
L14             1 S L13 AND (CONTRAST? OR IMAG?)
L15           855 S  L1 AND CELL####(3A)TARGET?
L16           263 S RECEPTOR? AND L15
L17            12 S L16 AND L2
L18             8 S L2 AND CELL####(4A)UPTAKE?
L19             0 S L18 AND HYDROPHOB?
```

=> D L12 BIB ABS 1-13

L12 ANSWER 1 OF 13 HCAPLUS COPYRIGHT 1996 ACS
AN 1996:456098 HCAPLUS
DN 125:107063
TI **Cationic** amphiphiles and plasmids for intracellular **delivery** of therapeutic molecules
IN Siegel, Craig S.; Harris, David J.; Lee, Edward R.; Hubbard, Shirley C.; Cheng, Seng H.; Eastman, Simon J.; Marshall, John; Scheule, Ronald K.; Yew, Nelson S.; et al.
PA Genzyme Corporation, USA
SO PCT Int. Appl., 152 pp.
CODEN: PIXXD2
PI WO 9618372 A2 960620
DS W: AM, AT, AU, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IS, JP, KE, KG, KP, KR, KZ, LK, LR, LT, LU, LV, MD, MG, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ, TM, TT
RW: AT, BE, BF, BJ, CF, CG, CH, CI, CM, DE, DK, ES, FR, GA, GB, GR, IE, IT, LU, MC, ML, MR, NE, NL, PT, SE, SN, TD, TG
AI WO 95-US16174 951208
PRAI US 94-352479 941209
US 95-540867 951011
US 95-545473 951019
DT Patent
LA English
OS MARPAT 125:107063
AB Novel **cationic** amphiphiles are provided that facilitate transport of biol. active (therapeutic) mols. into cells. The amphiphiles contain lipophilic groups derived from steroids, from mono or dialkylamines, or from alkyl or acyl groups; and **cationic** groups, protonatable at physiol. pH, derived from amines, alkylamines or polyalkylamines. Thus, N4-**spermidine** cholesteryl carbamate provided an .apprx.20-fold enhancement of the transfection ability of plasmid pCMVHI-CAT (chloramphenicol acetyltransferase-encoding) in mice. There are provided also therapeutic compns. prepd. typically by contacting a dispersion of one or more **cationic** amphiphiles with the therapeutic mols. Therapeutic mols. that can be **delivered** into cells according to the practice of the invention include **DNA**, RNA, and polypeptides. Representative uses of the therapeutic compns. of the invention include providing gene therapy, and **delivery** of antisense **polynucleotides** of biol. active polypeptides to cells. With respect to therapeutic compns. for gene therapy, the **DNA** is provided typically in the form of a plasmid for complexing with the **cationic** amphiphile. Novel and highly effective plasmid constructs are also disclosed, including those that are particularly effective at providing gene therapy for clin. conditions complicated by inflammation. Several vectors were constructed for improved **delivery** of the gene the cystic fibrosis transmembrane conductance regulator (CFTR) under control of the human cytomegalovirus promoter/enhancer during **cationic** amphiphile-mediated gene transfer. Addnl., targeting of organs for gene therapy by i.v. administration of therapeutic compns. is described. Syntheses are described for N4-spermine cholesteryl carbamate, N4-(N'-cholesteryl carbamate glycineamide)-spermine, N4-

spermidine-2,3-dilauryloxypropylamine, and N4-spermine-2,3-dilauryloxypropylamine.

L12 ANSWER 2 OF 13 HCAPLUS COPYRIGHT 1996 ACS
AN 1996:404762 HCAPLUS
DN 125:67763
TI Cell-specific gene **delivery** vehicles for **delivery** of paramagnetic ions
IN Kayyem, Jon F.; Meade, Thomas J.; Fraser, Scott E.
PA California Institute of Technology, USA
SO PCT Int. Appl., 37 pp.
CODEN: PIXXD2
PI WO 9611712 A2 960425
DS W: AL, AM, AT, AU, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IS, JP, KE, KG, KP, KR, KZ, LK, LR, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ
RW: AT, BE, BF, BJ, CF, CG, CH, CI, CM, DE, DK, ES, FR, GA, GB, GR, IE, IT, LU, MC, ML, MR, NE, NL, PT, SE, SN, TD, TG
AI WO 95-US14621 951011
PRAI US 94-321552 941012
DT Patent
LA English
AB A **delivery** vehicle is described that is capable of being specifically bound to and taken into **targeted cells**, **delivering** numerous physiol. agents, particularly paramagnetic ions for magnetic resonance imaging (MRI) of the cells. The **delivery** vehicle comprises a **polymeric** mol. having a net **pos**. charge complexed with another **polymeric** mol. having a net **neg**. charge. **Cell targeting** moieties and physiol. agents, including contrast agents and therapeutic agents, are attached to one or both of the **polymeric** mols. In one embodiment, the **polymeric** mol. having a net **neg**. charge is a **nucleic** acid. Thus, the **delivery** vehicles can be used in clin. protocols in which **nucleic** acids for gene therapy and agents for MRI contrast are co-transported to specific cells allowing medical imaging monitoring of **nucleic** acid **delivery**. A suspension of K562 cells were added to a complex of gadolinium-diethylenetriaminepentaacetic acid-polyD-lysine-**DNA** /transferrin (prepn. given) and allowed to incubate for 10 h at 37.degree.. The controls were simultaneously treated with free transferrin to competitively inhibit the receptor mediated uptake of MRI contrast agent **delivery** vehicle. MRI images of the cells transfected with particles contg. gadolinium-diethylenetriaminepentaacetic acid-**poly**-D-**lysine** showed intense signal indicative of gadolinium contrast enhancement, while the addn. of free transferrin competitively inhibited the uptake of the particles and reduced the MRI contrast.

L12 ANSWER 3 OF 13 HCAPLUS COPYRIGHT 1996 ACS
AN 1996:340828 HCAPLUS
DN 125:2971
TI **Delivery** of **nucleic** acids to cells for transfection using hypericin-**polyamine** complexes
IN Lavie, Gad; Prince, Alfred M.
PA New York University, USA; New York Blood Center

SO PCT Int. Appl., 47 pp.
CODEN: PIXXD2
PI WO 9607731 A1 960314
DS W: AU, CA, JP
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE
AI WO 95-US11709 950905
PRAI US 94-300725 940902
DT Patent
LA English
OS MARPAT 125:2971
AB A method for transfection of cultured mammalian cell is provided. The cell is contacted with a complex of the **nucleic** acid with a hydrophobic, membrane-binding **anion** and a **polycation**. The hydrophobic **anion** may comprise a polycyclic arom. dione (such a hypericin or its analogs), an anthraquinone, an emodin anthrone deriv., a cercosporine deriv., or a fatty acid; the **polycation** may comprise **polylysine**, polyarginine, polyasparagine, or various polyalkyleneamines. Thus, a 36-mer oligodeoxyribonucleotide forms a complex with **polylysine** and hypericin. The complex is 40-50% assocd. with murine T-lymphoblastoid cells, whereas only .apprx.1% is assocd. when **DNA** was added to the cells in the absence of hypericin or **polylysine**. HIV p55 gag expression was inhibited in CEM cell cultures exposed to an antisense phosphorothioate **oligonucleotide** complexed with hypericin and **polylysine**, whereas the **oligonucleotide** alone, hypericin alone, and **polylysine** alone were relatively ineffective.

L12 ANSWER 4 OF 13 HCAPLUS COPYRIGHT 1996 ACS
AN 1996:212906 HCAPLUS
DN 124:252118
TI Folate-targeted, **anionic** liposome-entrapped **polylysine**-condensed **DNA** for tumor cell-specific gene transfer
AU Lee, Robert J.; Huang, Leaf
CS Lab. Drug Targeting, Dep. Pharm., Univ. Pittsburgh Sch. Med., Pittsburgh, PA, 15261, USA
SO J. Biol. Chem. (1996), 271(14), 8481-7
CODEN: JBCHA3; ISSN: 0021-9258
DT Journal
LA English
AB We have developed a lipidic gene transfer vector, LPDII, where **DNA** was first complexed to **polylysine** at a ratio of 1:0.75 (wt./wt.) and then entrapped into folate-targeted pH-sensitive **anionic** liposomes composed of dioleoyl phosphatidylethanolamine (DOPE)/cholesteryl hemisuccinate/folate-polyethylene glycol-DOPE (6:4:0.01 mol/mol) via charge interaction. LPDII transfection of KB cells, a cell line overexpressing the tumor marker folate receptor, was affected by both the lipid to **DNA** ratio and the lipid compn. At low lipid to **DNA** ratios (e.g. 4 and 6), LPDII particles were **pos**. charged; transfection and cellular uptake levels were independent of the folate receptor and did not require a pH-sensitive lipid compn. Meanwhile, transfection and uptake of **neg**. charged LPDII particles, i.e. those with high lipid to **DNA** ratios (e.g. 10 and 12), were folate receptor-dependent and required a pH-sensitive lipid compn. The transfection activity of LPDII was

lost when the inverted cone-shaped DOPE was replaced by dioleoyl phosphatidylcholine. LPDII particles with lipid to **DNA** ratios of 4, 6, 10, and 12 were .apprx.20-30 times more active than **DNA**.cntdot.3-.beta.-[N-(N',N'-dimethylethane)carbamoyl]chole sterol **cationic** liposome complexes in KB cells and were much less cytotoxic. On the sucrose gradient, LPDII particles had a migration rate in between those of the free **DNA** and the **DNA**.cntdot.**polylysine** complex. An electron micrograph of LPDII showed a structure of spherical particles with a **pos**. stained core enclosed in a lipidic envelope with a mean diam. of 74 .+-. 14 nm. This novel gene transfer vector may potentially be useful in gene therapy for tumor-specific **delivery**.

L12 ANSWER 5 OF 13 HCAPLUS COPYRIGHT 1996 ACS
AN 1996:15602 HCAPLUS
DN 124:105484
TI Potentiation of **Cationic** Liposome-Mediated Gene **Delivery** by **Polycations**
AU Gao, Xiang; Huang, Leaf
CS School of Medicine, University of Pittsburgh, Pittsburgh, PA, 15261, USA
SO Biochemistry (1996), 35(3), 1027-36
CODEN: BICHAW; ISSN: 0006-2960
DT Journal
LA English
OS CJACS-IMAGE; CJACS
AB We discovered that several high mol. wt. **cationic** polymers, such as **poly**(L-**lysine**) and protamine, can enhance the transfection efficiency of several types of **cationic** liposomes by 2-28-fold in a no. of cell lines in vitro. Small **polycations** such as spermine and a **cationic** decapeptide derived from SV40 T-antigen were only moderately active. The addn. of **poly**(L-**lysine**) and protamine dramatically reduced the particle size of the complex formed between **DNA** and **cationic** liposomes and rendered **DNA** resistant to the nuclease activity. The complexes composed of **DNA**, **poly**(L-**lysine**), and **cationic** lipids were purified from an excess of free liposomes with sucrose gradient ultracentrifugation. Purified complex formed at low **cationic** liposome ratio was poor in lipid content and only had weak transfection activity. Addn. of free liposome to the purified complex significantly enhanced the transfection activity. In contrast, complexes formed at a higher initial ratio of liposome to **DNA** had a higher lipid content and were highly active in transfection; the activity was about 3-9-fold more active than the corresponding complex before purifn. **Neg**. stain EM studies revealed that the most active complexes prepd. from 40 nmol of lipid, 0.5 .mu.g of **poly**(L-**lysine**), and 1 .mu.g of **DNA** and purified by gradient ultracentrifugation were spherical, electron dense, small (<100 nm in diam.) particles, and some of them were assocd. with lipid membranes. These highly active, stable, small-sized lipid/**poly**(L-**lysine**)/**DNA** complexes represent a new class of nonviral gene **delivery** vehicles that might be useful in gene therapy.

L12 ANSWER 6 OF 13 HCAPLUS COPYRIGHT 1996 ACS

AN 1995:924529 HCAPLUS
TI Reversible attachment of effectors to plasmid **DNA** for gene therapy.
AU Wyman, Tara B.; Plank, Christian; Szoka, Francis C. Jr.
CS School Pharmacy, University California, San Francisco, CA, 94143-0446, USA
SO Book of Abstracts, 210th ACS National Meeting, Chicago, IL, August 20-24 (1995), Issue Pt. 2, NUCL-008 Publisher: American Chemical Society, Washington, D. C.
CODEN: 61XGAC
DT Conference; Meeting Abstract
LA English
AB The advent of gene therapy has catalyzed the search for safe and effective **DNA** trnasfection systems. The actual and perceived safety issues surrounding viral vectors has turned attention to synthetic vectors for gene **delivery**. Most of the approaches to synthetic vectors use the **DNA** plasmid as a scaffold and have assocd. multiple components with the **DNA**, e.g., targeting ligands, condensing agents and membrane destabilization moieties, to obtain effective **delivery**. In the **target cell**, the effectors must come off the **DNA** to obtain effective gene expresion. The effector mols. are coupled to groups that can bind to **DNA** through non-covalent interactions. A variety of forces have been used to attached the effectors to the **neg**. charged **DNA** including electrostatic interactions, e.g. **polylysine** or **cationic** dendrimer tails, intercalation via acridine groups, hydrogen bonding via the minor grove of **DNA**(distamycin analogs), hydrophobic interactions via lipid groups or a combination of hydrogen bonding and base stacking via triplex formation. The physico-chem. characteristics and relative merits of each approach will be discussed and examples will be presented.

L12 ANSWER 7 OF 13 HCAPLUS COPYRIGHT 1996 ACS
AN 1995:736555 HCAPLUS
DN 123:160579
TI A versatile vector for gene and **oligonucleotide** transfer into cells in culture and in vivo: polyethylenimine
AU Boussif, Otmane; Lezoualc'h, Frank; Zanta, Maria Antonietta; Mergny, Mojgan Djavaheri; Scherman, Daniel; Demeneix, Barbara; Behr, Jean-Paul
CS Lab. Chim. Genetique, Unite Recherche Associee 1386, Cent. Natl. Recherche Scientifique, Fac. Pharmacie, Illkirch, F-67401, Fr.
SO Proc. Natl. Acad. Sci. U. S. A. (1995), 92(16), 7297-301
CODEN: PNASA6; ISSN: 0027-8424
DT Journal
LA English
AB Several **polycations** possessing substantial buffering capacity below physiol. pH, such as lipopolyamines and polyamidoamine **polymers**, are efficient transfection agents per se-i.e., without the addn. of **cell targeting** or membrane-disruption agents. This observation led the authors' to test the **cationic polymer** polyethylenimine (PEI) for its gene-**delivery** potential. Indeed, every third atom of PEI is a protonable amino nitrogen atom, which makes the **polymeric** network an effective "proton sponge" at virtually any pH. Luciferase reporter gene transfer with this **polycation** into a variety of cell lines and primary cells

gave results comparable to, or even better than, lipopolyamines. Cytotoxicity was low and seen only at concns. well above those required for optimal transfection. **Delivery** of **oligonucleotides** into embryonic neurons was followed by using a fluorescent probe. Virtually all neurons showed nuclear labeling, with no toxic effects. The optimal PEI **cation**/**anion** balance for in vitro transfection is only slightly on the **cationic** side, which is advantageous for in vivo **delivery**. Indeed, intracerebral luciferase gene transfer into newborn mice gave results comparable (for a given amt. of **DNA**) to the in vitro transfection of primary rat brain endothelial cells or chicken embryonic neurons. Together, these properties make PEI a promising vector for gene therapy and an outstanding core for the design of more sophisticated devices. The hypothesis is that its efficiency relies on extensive lysosome buffering that protects **DNA** from nuclease degrdn., and consequent lysosomal swelling and rupture that provide an escape mechanism for the PEI/**DNA** particles.

L12 ANSWER 8 OF 13 HCAPLUS COPYRIGHT 1996 ACS
AN 1995:666220 HCAPLUS
DN 123:81263
TI Natural killer (NK) activity in human responders and nonresponders to stimulation by anti-CD16 antibodies
AU Galactiuc, Cecilia; Gherman, Maria; Metes, Diana; Sulica, A.; DeLeo, A.; Whiteside, Theresa L.; Heberman, R. B.
CS Center for Immunology, Bucharest, Rom.
SO Cell. Immunol. (1995), 163(2), 167-77
CODEN: CLIMB8; ISSN: 0008-8749
DT Journal
LA English
AB Various anti-Fc.gamma.RIII (CD16) monoclonal antibodies (mAbs) are shown here to have **pos**. or **neg**. modulatory effects on human NK cells. Thus, 3G8 mAb (IgG1) triggered a dose-dependent augmentation of NK activity in 67% (23/34) of individuals tested, who were designated as responders. All four IgG1 anti-CD16 mAb tested (BL-LGL/1, B73.1, Leu11c, and 3G8) were stimulatory for NK cells isolated from responders, whereas six non-IgG1 anti-CD16 mAbs were either inhibitory or had no significant effects on NK activity. The up-regulation of NK activity in responders was not attributable to an increase in either the conjugate formation or the **delivery** of the lethal hit to **target cells**. This mAb-mediated up-regulation of NK activity was shown to be assocd. with a recycling capacity higher than that of controls and with enhanced release of cytokines by activated NK cells. Anti-CD16 mAb inhibited binding of either monomeric or **polymeric** IgG to Fc.gamma.RIIIA on NK cells. Also, mAb 3G8 or its F(ab')2 fragments decreased or reversed inhibition of NK activity induced by monomeric IgG (mIgG). The data indicate that regulation of NK activity via the Fc.gamma.RIIIA is influenced by dose-dependent interactions between cytophilic mIgG and anti-CD16 mAb of IgG1 isotype.

L12 ANSWER 9 OF 13 HCAPLUS COPYRIGHT 1996 ACS
AN 1995:238169 HCAPLUS
DN 122:38716
TI Interaction between charged peptides and **nucleic** acids: development of a histone- or peptide-mediated potential drug

delivery system
AU Wada, Akira; Suzuki, Yosuke; Okayama, Minenobu; Sato, Shuji; Shimayama, Takashi; Oya, Masanao; Uchida, Chieko; Koguma, Tetsuhiko; Nishikawa, Satoshi; et al.
CS National Inst. Biosci. Human Technology, AIST, 305, Japan
SO Nucleic Acids Symp. Ser. (1994), 31(21st Symposium on Nucleic Acids Chemistry, 1994), 227-8
CODEN: NACSD8; ISSN: 0261-3166
DT Journal
LA English
AB As a step toward the goal to develop a highly efficient **nucleic** acid **delivery** system, that might facilitate receptor-mediated endocytosis of **DNA**/RNA macromols. into cells, we examd. interactions between **pos**. charged polypeptides and **neg**. charged **nucleic** acids. Poly-**cationic** amino acids used in this study included **poly**-L-**Lysine**, **poly**-L-(**Lysine**:Serine) random copolymer, poly(D,L)-Lysine random copolymer, and histone. Ribozymes and antisense **oligonucleotides**, that are thought to be interesting tools for selective inhibition of gene expression, were found to form complexes with poly-**cationic** amino acids except the poly(D,L)-Lysine random copolymer. Such complexes were much more resistant to degrdn. by nucleases in human serum. Com. available histone may be used as a carrier for antisense **DNA** and plasmids. However, it was not a suitable carrier for ribozymes or antisense RNA because it was contaminated with significant amt. of RNases. Available data suggest that **oligonucleotide**:poly-**cationic** amino acid complexes have high potential as carriers for **oligonucleotide** drugs.

L12 ANSWER 10 OF 13 HCAPLUS COPYRIGHT 1996 ACS
AN 1995:197839 HCAPLUS
DN 122:64104
TI A total **delivery** system of genetically engineered drugs or cells for diseased vessels: concept, materials, and fabricated prototype device
AU Kito, Hiroyuki; Suzuki, Fumiaki; Nagahara, Shunji; Nakayama, Yasuhide; Tsutsui, Yoko; Tsutsui, Nobumasa; Nakajima, Nobuyuki; Matsuda, Takehisa
CS Department of Bioengineering, National Cardiovascular Center Research Institute, Suita, 565, Japan
SO ASAIO J. (1994), 40(3), M260-M266
CODEN: AJOUET; ISSN: 1058-2916
DT Journal
LA English
AB The development of a percutaneous procedure using a catheterized system for diseased vessels has been increasingly in demand in conjunction with gene therapy using genetically engineered drugs (antisense) and cells. The authors' strategic concept realizes revascularization at narrowed, diseased sites and **delivery** of drugs or cells into the diseased tissues or **targeted** **cells**. An inflatable, drug-releasing double balloon is installed at the tip of a catheter. The outer balloon, fabricated with micropores (diams. of 20 and 30 mm) by an excimer laser ablation technique, releases a viscous soln. contg. a photoreactive **polymer** and drug or cells on inflation of the inner balloon. A photoresponsive water-sol. **polymer**, molecularly designed

for its ability to achieve prolonged local residency of antisense **DNA** at the tissue level and enhanced transmembrane transport at the cellular level, is premixed with antisense **oligonucleotide** drug. On light irradn., the nonionic **polymer** is reversibly converted to a **pos**. charged **polymer** that can be complexed with highly **neg**. charged antisense **DNA** (c-myb), which may enhance the transmembrane **delivery** of antisense. On cessation of irradn., the complex slowly dissocs. to function intracellularly as an antisense drug, resulting in inhibition of cell proliferation. Thus, our integrated, dual-function balloon system may contribute to mech. dilatation gene therapies at diseased vessels.

L12 ANSWER 11 OF 13 HCAPLUS COPYRIGHT 1996 ACS
AN 1993:415334 HCAPLUS
DN 119:15334
TI Targeted **delivery** of poly- or **oligonucleotides** to cells
IN Wu, George Y.; Wu, Catherine H.
PA University of Connecticut, USA
SO PCT Int. Appl., 44 pp.
CODEN: PIXXD2
PI WO 9304701 A1 930318
DS W: AT, AU, BB, BG, BR, CA, CH, CS, DE, DK, ES, FI, GB, HU, JP, KP, KR, LK, LU, MG, MN, MW, NL, NO, PL, RO, RU, SD, SE
RW: AT, BE, BF, BJ, CF, CG, CH, CI, CM, DE, DK, ES, FR, GA, GB, GR, IE, IT, LU, MC, ML, MR, NL, SE, SN, TD, TG
AI WO 92-US7339 920904
PRAI US 91-755083 910905
US 91-788119 911104
US 92-864003 920403
DT Patent
LA English
AB A single-stranded poly- or **oligonucleotide**, e.g. an antisense **oligonucleotide** or ribozyme, is complexed to a conjugate of a cell-specific binding agent (e.g. a receptor-specific structure which mediates internalization of the complex) with a poly- or **oligonucleotide**-binding agent for **targeting** to a specific **cell**(s) to block expression of a gene(s) therein. The poly- or **oligonucleotide**-binding agent is e.g. a **polycationic** protein which stably complexes the **oligonucleotide** under extracellular conditions and releases it under intracellular conditions so that it can hybridize with the target RNA. The **oligonucleotide** can be directed against cellular genes (e.g. cellular oncogenes) or genes of noncellular origin (e.g. viral oncogenes or genes of a pathogen). Thus, asialoorosomucoid was conjugated with **poly**-L-**lysine** using 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide and then complexed with a 21-mer oligodeoxynucleotide complementary to a portion of human hepatitis B virus synthesized with phosphorothioate linkages and end-labeled with 32P. Uptake of the complex by human HepG2 hepatoma cells, which were **pos**. for asialoglycoprotein receptors, was 12-fold faster than uptake of antisense **DNA** alone or uptake of the complex by receptor-**neg**. cells, and inhibited expression of the hepatitis B virus surface antigen gene by the cells by .apprx.80%. The complex was also taken up specifically by the liver of rats in vivo.

L12 ANSWER 12 OF 13 HCAPLUS COPYRIGHT 1996 ACS
AN 1993:227392 HCAPLUS
DN 118:227392
TI Targeted transfection and expression of hepatitis B viral **DNA** in human hepatoma cells
AU Liang, T. Jake; Makdisi, Walid J.; Sun, Susan; Hasegawa, Kiyoshi; Zhang, Ying; Wands, Jack R.; Wu, Catherine H.; Wu, George Y.
CS Cancer Cent., Massachusetts Gen. Hosp., Boston, MA, 02114, USA
SO J. Clin. Invest. (1993), 91(3), 1241-6
CODEN: JCINAO; ISSN: 0021-9738
DT Journal
LA English
AB A sol. **DNA** carrier system consisting of an asialoglycoprotein covalently linked to **poly**-L-**lysine** was used to bind **DNA** and **deliver** hepatitis B virus (HBV) **DNA** constructs to asialoglycoprotein receptor-**pos**. human hepatoma cells. Four days after transfection with surface or core gene expression constructs, HBsAg and HBeAg in the media were measured to be 16 ng/mL and 32 U/mL per 107 cells, resp. Antigen prodn. was completely inhibited by the addn. of an excess of asialoorosomucoid. On the other hand, asialoglycoprotein receptor-**neg**. human hepatoma cells, SK-Hep1, did not produce any viral antigens under identical conditions after incubation with HBV **DNA** complexed to a conjugate composed of asialoorosomucoid and **poly**-L-**lysine**. Using a complete HBV genome construct, HBsAg and HBeAg levels reached 16 ng/mL and 16 U/mL per 107 cells, resp. Northern blots revealed characteristic HBV RNA transcripts including 3.5-, 2.4-, and 2.1-kb fragments. Intracellular and extracellular HBV **DNA** sequences including relaxed circular, linear and single stranded forms were detected by Southern blot hybridization. Finally, 42-nm Dane particles purified from the spent culture medium were visualized by electron microscopy. Evidently, a targetable **DNA** carrier system can transfect HBV **DNA** in vitro resulting in the prodn. of complete HBV virions.

L12 ANSWER 13 OF 13 HCAPLUS COPYRIGHT 1996 ACS
AN 1992:221370 HCAPLUS
DN 116:221370
TI Targeted **delivery** of **DNA** by liposomes and **polymers**
AU Zhou, Xiaohuai; Huang, Leaf
CS Dep. Biochem., Univ. Tennessee, Knoxville, TN, 37996-0840, USA
SO J. Controlled Release (1992), 19(1-3), 269-74
CODEN: JCREEC; ISSN: 0168-3659
DT Journal; General Review
LA English
AB A review with 17 refs. **Cationic** quaternary ammonium detergents stabilized the lamellar phase of dioleoylphosphatidylethanolamine (DOPE) to form liposomes. Such **cationic** liposomes interacted with and **delivered** **DNA** to tissue culture cells with high efficiency. **Pos**. charged **polymers** conjugated with lipids were also active in **DNA delivery**. The transfection activity of the lipopoly-L-lysine under optimal conditions was approx. 3-fold higher than that of Lipofectin, a com. available

liposome reagent. Moreover, the conjugate was much more resistant to serum neutralizing effect. Specific **delivery** of **DNA** to **target cells** was achieved by using the **anionic** pH-sensitive immunoliposomes. It was found that both pH-sensitivity and incorporation of antibody on liposome surface were important for the high transfection activity. Both the short- and long-term transformation efficiency had been detd. and shown to be more superior to those of traditional calcium phosphate pptn. method. Successful **delivery** and expression of the exogenous genes mediated by pH-sensitive immunoliposomes had been demonstrated in both tissue cultured cells and in a mouse model.

=> D L14 ALL

L14 ANSWER 1 OF 1 HCAPLUS COPYRIGHT 1996 ACS
AN 1996:404762 HCAPLUS
DN 125:67763
TI Cell-specific gene **delivery** vehicles for **delivery** of paramagnetic ions
IN Kayyem, Jon F.; Meade, Thomas J.; Fraser, Scott E.
PA California Institute of Technology, USA
SO PCT Int. Appl., 37 pp.
CODEN: PIXXD2
PI WO 9611712 A2 960425
DS W: AL, AM, AT, AU, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IS, JP, KE, KG, KP, KR, KZ, LK, LR, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ
RW: AT, BE, BF, BJ, CF, CG, CH, CI, CM, DE, DK, ES, FR, GA, GB, GR, IE, IT, LU, MC, ML, MR, NE, NL, PT, SE, SN, TD, TG
AI WO 95-US14621 951011
PRAI US 94-321552 941012
DT Patent
LA English
IC ICM A61K047-48
ICS A61K049-00
CC 63-6 (Pharmaceuticals)
Section cross-reference(s): 8
AB A **delivery** vehicle is described that is capable of being specifically bound to and taken into **targeted cells**, **delivering** numerous physiol. agents, particularly paramagnetic ions for magnetic resonance **imaging** (MRI) of the cells. The **delivery** vehicle comprises a polymeric mol. having a net **pos**. charge complexed with another polymeric mol. having a net **neg**. charge. **Cell targeting** moieties and physiol. agents, including **contrast** agents and therapeutic agents, are attached to one or both of the polymeric mols. In one embodiment, the polymeric mol. having a net **neg**. charge is a **nucleic** acid. Thus, the **delivery** vehicles can be used in clin. protocols in which **nucleic** acids for gene therapy and agents for MRI **contrast** are co-transported to specific cells allowing medical **imaging** monitoring of **nucleic** acid **delivery**. A suspension of K562 cells were added to a complex of gadolinium-diethylenetriaminepentaacetic acid-polyD-lysine-**DNA**/transferrin (prepn. given) and allowed to incubate for 10 h at 37.degree.. The controls were

simultaneously treated with free transferrin to competitively inhibit the receptor mediated uptake of MRI **contrast** agent **delivery** vehicle. MRI **images** of the cells transfected with particles contg. gadolinium-diethylenetriaminepentaacetic acid-poly-D-lysine showed intense signal indicative of gadolinium **contrast** enhancement, while the addn. of free transferrin competitively inhibited the uptake of the particles and reduced the MRI **contrast**.

ST gene **delivery** vehicle paramagnetic ion; magnetic resonance **imaging** cell **delivery**

IT Neoplasm inhibitors
Therapeutics
(cell-specific gene **delivery** vehicles for **delivery** of paramagnetic ions)

IT Deoxyribonucleic acids
Transferrins
RL: RCT (Reactant)
(cell-specific gene **delivery** vehicles for **delivery** of paramagnetic ions)

IT Polymers, biological studies
RL: THU (Therapeutic use); BIOL (Biological study); USES (Uses)
(cell-specific gene **delivery** vehicles for **delivery** of paramagnetic ions)

IT **Imaging**
(NMR, cell-specific gene **delivery** vehicles for **delivery** of paramagnetic ions)

IT **Imaging**
(**contrast** agents, cell-specific gene **delivery** vehicles for **delivery** of paramagnetic ions)

IT Amines, biological studies
RL: THU (Therapeutic use); BIOL (Biological study); USES (Uses)
(poly-, cell-specific gene **delivery** vehicles for **delivery** of paramagnetic ions)

IT 67-43-6, Diethylenetriaminepentaacetic acid 7790-28-5, Sodium periodate 10138-52-0, Gadolinium trichloride 25104-18-1, Poly-L-lysine 25104-18-1D, Poly-L-lysine, conjugates with transferrins 38000-06-5, Poly-L-lysine 38000-06-5D, Poly-L-lysine, conjugates with transferrins
RL: RCT (Reactant)
(cell-specific gene **delivery** vehicles for **delivery** of paramagnetic ions)

IT 67-43-6DP, DTPA, gadolinium and poly-D-lysine complexes 67-43-6DP, Diethylenetriaminepentaacetic acid, reaction products with polylysine 7440-54-2DP, Gadolinium, DTPA and poly-D-lysine complexes 26853-89-4DP, Poly-D-lysine, gadolinium and DTPA complexes 26913-90-6DP, Poly-D-lysine, gadolinium and DTPA complexes
RL: RCT (Reactant); SPN (Synthetic preparation); PREP (Preparation)
(cell-specific gene **delivery** vehicles for **delivery** of paramagnetic ions)

IT 124-20-9, Spermidine 9002-06-6, Thymidine kinase 60239-18-1, Dota
RL: THU (Therapeutic use); BIOL (Biological study); USES (Uses)
(cell-specific gene **delivery** vehicles for **delivery** of paramagnetic ions)

=> D HIS

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     (FILE 'HCAPLUS' ENTERED AT 07:20:14 ON 25 SEP 96)
                DEL HIS   Y
L1          48268 S DELIVER?
L2            622 S L1 AND (POS OR POSITIV? OR POLYCATION? OR CATION?) AND
L3             17 S L2 AND (POLYMAINE# OR SPERMIDINE? OR POLYLYSINE OR POLY
L4             52 S L2 AND (POLYNUCLEO? OR DNA OR NUCLEIC OR OLIGONUCLEO?)
L5              5 S L2 AND POLYAMINE?
L6             19 S L3 OR L5
L7              9 S L6 AND L4
L8             21 S L2 AND CELL####(3A)TARGET?
L9              6 S L8 AND POLYMER?
L10            13 S L7 OR L9
L11             7 S L8 AND (L3 OR L4 OR L5)
L12            13 S L7 OR L9 OR L11
L13             7 S L4 AND CELL####(3A)TARGET?
L14             1 S L13 AND (CONTRAST? OR IMAG?)
L15           855 S  L1 AND CELL####(3A)TARGET?
L16           263 S RECEPTOR? AND L15
L17            12 S L16 AND L2
L18             8 S L2 AND CELL####(4A)UPTAKE?
L19             0 S L18 AND HYDROPHOB?
L20             0 S L14 NOT L12
L21             0 S L13 NOT L12
L22             8 S L17 NOT L12

=>
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=> D 1-8 BIB ABS

L22 ANSWER 1 OF 8 HCAPLUS COPYRIGHT 1996 ACS
AN 1995:735072 HCAPLUS
DN 123:141005
TI Hanging in the balance: natural killer **cell** recognition of **target cells**
AU Chambers, William H.; Brissette-Storkus, Cynthia S.
CS Pittsburgh Cancer Inst., Pittsburgh, PA, 15213, USA
SO Chem. Biol. (1995), 2(7), 429-35
CODEN: CBOLE2; ISSN: 1074-5521
DT Journal; General Review
LA English
AB A review with 60 refs. Natural killer (NK) cells kill certain tumor cells and virus-infected cells directly. Until recently, little was known about how they recognize their targets. Now, several candidate NK **receptors** have been identified, some of which may have carbohydrate ligands. Some of the **receptors** **deliver pos**. signals, others **neg**. signals. Thus NK cells seem to balance many different inputs to decide whether to kill a target. Substances discussed include Ly49; p58/NKATs; CD94; class I antigens; NK-TR1; 2B4; and p38.

L22 ANSWER 2 OF 8 HCAPLUS COPYRIGHT 1996 ACS
AN 1994:571340 HCAPLUS
DN 121:171340
TI Epidermal growth factor **receptors** in human breast carcinoma **cells**: a potential selective **target** for transforming growth factor .alpha.-Pseudomonas exotoxin 40 fusion protein
AU Arteaga, Carlos L.; Hurd, Stephen D.; Dugger, Teresa C.; Winnier, Angela R.; Robertson, J. Bruce
CS Sch. Med., Vanderbilt Univ., Nashville, TN, 37232, USA
SO Cancer Res. (1994), 54(17), 4703-9
CODEN: CNREA8; ISSN: 0008-5472
DT Journal
LA English
AB Epidermal growth factor (EGF) **receptors** are expressed in high levels by some poor prognosis breast tumors. We have examd. the cytotoxic effect of the tumor growth factor .alpha. (TGF.alpha.)-.DELTA.Cys-Pseudomonas exotoxin (PE40) recombinant fusion protein on normal and tumorigenic human breast epithelial cells in vitro and in vivo. The MDA-468, MDA-231, BT-20, and MCF-7AER estrogen **receptor-neg**., EGF **receptor**-rich breast cancer lines were exquisitely sensitive in vitro to TGF.alpha.-.DELTA.Cys-PE40 with a 50% inhibitory concn. of .ltoreq.0.02 nM. The estrogen **receptor-pos**., low EGF **receptor** MCF-7, ZR75-1, and T47D cells were less sensitive to the fusion toxin with a 50% inhibitory concn. of >0.2 nM. The nontumorigenic cell lines 184, 184A1, and 184B5 were relatively resistant to TGF.alpha.-.DELTA.Cys-PE40 despite exhibiting high levels of EGF **receptors**. Continuous i.p. administration of TGF.alpha.-.DELTA.Cys-PE40 via an osmotic minipump at a dose of 0.4 .mu.g/g/day over 7 days inhibited MZDA-468, MA-231, and BT-20 but not MCF-7 tumor growth in female athymic mice. Host tissue toxicity was not obsd. with this dose of TGF.alpha.-.DELTA.Cys-PE40. Mixed MDA-468/MCF-7 tumors were established in

nude mice after coinoculation of both cell types in estrogen-supplemented animals. EGF **receptor** immunohistochem. and immunoblot procedures indicated that TGF .alpha.-PE40 eliminated the MDA-468 cells while sparing the adjacent MCF-7 cells. By immunoblot, EGF **receptors** were consistently more abundant in tumor tissue than in adjacent nontumor tissue from the same mastectomy specimen. These data support the notion that EGF **receptors** can be selectively targeted in human breast cancer cells for the **delivery** of antitumor agents. Further clin. studies with TGF-.alpha.-.DELTA.Cys-PE40 and other chimeric toxins using the same **cellular target** will address this possibility.

L22 ANSWER 3 OF 8 HCAPLUS COPYRIGHT 1996 ACS
AN 1994:214901 HCAPLUS
DN 120:214901
TI Targeting of T lymphocytes against EGF-**receptor**+ tumor cells by bispecific monoclonal antibodies: requirement of CD3 molecule crosslinking for T-cell activation
AU Ferrini, Silvano; Cambiaggi, Anna; Sforzini, Sabrina; Marciano, Sabrina; Canevari, Silvana; Mezzanzanica, Delia; Colnaghi, Maria Ines; Grossi, Carlo Enrico; Moretta, Lorenzo
CS Ist. Naz. Ric. Cancro, Genoa, Italy
SO Int. J. Cancer (1993), 55(6), 931-7
CODEN: IJCNAW; ISSN: 0020-7136
DT Journal
LA English
AB Targeting of T lymphocytes against epidermal growth-factor-**receptor** (EGF-R)+ tumor cells was achieved by constructing a hybrid hybridoma which secretes an anti-EGF-R/anti-CD3 bispecific monoclonal antibody (biMAb) of hybrid isotype (IgG1/IgG2a). Purifn. of biMAb mols. from parental anti-EGF-R and anti-CD3 MAbs was performed by protein-A chromatog. The purified biMAb was able to trigger the lysis of EGF-R+ tumor cell lines (A431, IGROV-1, MDA-468, and U-87) and of NIH-3T3 transfectants expressing human EGF-R by cytolytic T lymphocytes, but it was ineffective in the case of EGF-R-**neg**. tumor targets. Normal EGF-R+ cells (keratinocytes and endometrial cells) were also susceptible to biMAb-targeted cytolysis. However, the amt. of biMAb required to induce half-maximal cytolysis of tumor cells overexpressing the EGF-R mol. (A431) was considerably lower than that required to induce lysis of EGF-R+ tumor or normal cells which express EGF-R at considerably lower d. The ability of such biMAbs to **deliver** activation signals to T cells was evaluated by Ca2+ mobilization and lymphokine prodn. expts. The sol. anti-EGF-R/anti-CD3 biMAb failed to induce intracellular Ca2+ increases, which occurred only after crosslinking induced by an anti-mouse IgG antibody. Secretion of lymphokines (IFN-.gamma., TNF-.alpha., and GM-CSF) was induced by contact of the biMAb-coated effector cells with the relevant tumor target, whereas the sol. biMAb was virtually ineffective. In addn., biMAb-coated effector cells retained the ability to recognize and to lyse EGF-R+ tumor cells for a prolonged period of time. Thus, the activation of effector **cells targeted** by biMAbs can only occur at the tumor site, where crosslinking of surface CD3 mols. is induced by contact with the tumor cells.

L22 ANSWER 4 OF 8 HCAPLUS COPYRIGHT 1996 ACS
AN 1993:668790 HCAPLUS

DN 119:268790
TI Functional studies of adhesion molecules on CD4-CD8-double **negative** T cells of autoimmune MRL/Mp-lpr/mice
AU Wang, Weila
CS Inst. Immunol. Sci., Hokkaido Univ., Sapporo, 060, Japan
SO Hokkaido Igaku Zasshi (1993), 68(5), 755-66
CODEN: HOIZAK; ISSN: 0367-6102
DT Journal
LA Japanese
AB MRL/Mp-lpr/lpr (MRL-lpr) mice have been used for a model of human systemic lupus erythematosus. This strain of mice homozygous for an autosomal recessive mutation, lpr (lymphoproliferation), develops massive lymphadenopathy with the expansion of CD4-CD8-(double **neg**.; DN) T cells. Recently it was demonstrated that lpr mice have defects in the gene of Fas antigen which mediates apoptosis, indicating a possibility of defect in **neg**. selection of autoreactive T cells in the thymus of lpr mice. However, the mechanisms that control the accumulation of DN T cells in lymph nodes, and the involvement of DN T cells in the clin. manifestation of disease, have not been well understood. In this study, the expression of various cell adhesion mols. on lymphocytes from MRL-lpr mice was examd. The strong expression of CD44 antigen as well as heat stable antigen (HSA) on abnormal DN T cells of lymph nodes was characteristic in MRL-lpr mice. Furthmore, the accumulation of DN T cells in lymph nodes might result from augmented binding of lymphocytes to endothelial cell surface of lymph nodes, possibly due to the failure of Mel-14 antigen shedding from DN T cell surface. In addn., monoclonal antibodies reactive with cell adhesion mols. such as CD44, Mel-14, CD45R and HSA expressed on DN T cells, could trigger the lytic activity of DN T cells and redirected DN T cell-mediated lysis of Fc-**receptor** -**pos**. **target cells** (EL-4). In contrast to T cell **receptor** (TCR)-mediated cytotoxicity, this redirected cytotoxicity was not inhibited by anti-lymphocyte function assocd. antigen-1 (LFA-1) antibody. Thus, cell adhesion mols. may play a major role in **delivering** the transmembrane signal to DN T cells of MRL-lpr mice that trigger the lytic activity. It is likely that DN T cells of MRL-lpr mice induce tissue damages by the interaction with ligand on vascular endothelium or extracellular matrix in vivo.

L22 ANSWER 5 OF 8 HCAPLUS COPYRIGHT 1996 ACS
AN 1993:139386 HCAPLUS
DN 118:139386
TI Human melanoma targeting with .alpha.-MSH-melphalan conjugate
AU Ghanem, G. E.; Libert, A.; Arnould, R.; Vercammen, A.; Lejeune, F.
CS Jules Bordet Inst., Univ. Libre Brusselles, Brussels, Belg.
SO Melanoma Res. (1991), 1(2), 105-14
CODEN: MREEEH; ISSN: 0960-8931
DT Journal
LA English
AB A conjugate made of .alpha.-MSH as a drug carrier and melphalan has been designed in order to **target** human melanoma **cells**. Iodination of the .alpha.-MSH moiety led to a relatively stable tracer which could be easily sepd. and analyzed by reverse phase high pressure liq. chromatog. The conjugate was found to be unstable at neutral pH and a serious denaturation can take place at concns. exceeding 100 .mu.g/mL, esp. in plasma.

Receptor-mediated cytotoxicity has been shown by the use of cultured .alpha.-MSH **receptor pos./neg** . cells as well as in vivo B16 murine melanoma model. Body distribution and uptake of the labeled compd. were unaltered as compared to those of labeled free hormone. .alpha.-MSH **receptor** recognition properties also remained unchanged with a better apparent affinity of the conjugate probably due to the alkylating activity of melphalan itself. Using human melanoma dendritic cells expressing more than 10,000 .alpha.-MSH binding sites per cell as an in vitro model, we were able to demonstrate higher cytotoxicities as compared to melphalan-treated cells. In contrast, melanoma cells with low receptivity did not show higher cytotoxicity. P388D1 mouse plasmocytoma cells lacking **receptors** were much more sensitive to melphalan than the conjugate. This phenomenon appeared to be related with the no. of binding sites expressed at the time of the expt. as well as cell differentiation and the doubling time. Our findings strongly support the concept of a **receptor**-mediated cytotoxicity and may enable the in vivo melphalan **delivery** to target tissues to be increased, achieving an improvement of drug penetration inside melanoma cells.

L22 ANSWER 6 OF 8 HCAPLUS COPYRIGHT 1996 ACS
AN 1992:433703 HCAPLUS
DN 117:33703
TI Self-assembling ion pair drugs for tumor targeting
IN Rideout, Darryl C.
PA Scripps Research Institute, USA
SO PCT Int. Appl., 17 pp.
CODEN: PIXXD2
PI WO 9205803 A1 920416
DS W: CA, JP
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LU, NL, SE
AI WO 91-US7292 911004
PRAI US 90-592926 901004
DT Patent
LA English
AB Sol. forms of **pos**. and **neg**. ion components of an ion pair capable of formation in situ in a **targeted** organism, **cell** or tissue, are administered to make ion pair conjugate affective to modify the target. Ion pair using tetrachloroplatinate as a **neg**. ion and org. **cations** are disclosed. A 50% increase in life span of mice implanted with L1210 tumors was achieved when a mixt. of dequalinium chloride (I) and K tetrachloroplatinate (II) were injected on day 1, 3, and 5. The optimal dose of the I:II mixt. was 1:4.75.

L22 ANSWER 7 OF 8 HCAPLUS COPYRIGHT 1996 ACS
AN 1991:629925 HCAPLUS
DN 115:229925
TI T-cell-**receptor**-independent activation of cytolytic activity of cytotoxic T lymphocytes mediated through CD44 and gp90MEL-14
AU Seth, Aruna; Gote, Lisa; Nagarkatti, Mitzi; Nagarkatti, Prakash S.
CS Dep. Biol., Virginia Polytech. Inst. and State Univ., Blacksburg, VA, 24061, USA
SO Proc. Natl. Acad. Sci. U. S. A. (1991), 88(17), 7877-81
CODEN: PNASA6; ISSN: 0027-8424

DT Journal
LA English
AB CD44 is a transmembrane glycoprotein found on a variety of cells including those of myeloid and lymphoid origin. CD44 is highly conserved among various species and is involved in the homing of lymphocytes and monocytes to lymph nodes, Peyer's patches, and sites of inflammation. In the present study, it is demonstrated that monoclonal antibody (mAb) 9F3, directed against murine phagocytic glycoprotein 1 (CD44) expressed on cytotoxic T lymphocytes (CTLs), can trigger the lytic activity of CTLs and redirect CTL-mediated lysis to antigen-**neg**. Fc **receptor-pos**. **target cells**. Similar redirected lysis was also inducible using mAb MEL-14, directed against the lymphocyte homing **receptor** for endothelium (gp90MEL-14). The redirected lysis induced by mAbs 9F3 and MEL-14 was similar to that induced by mAbs against the .alpha..beta. T-cell **receptor** or CD3. In contrast, mAbs directed against CD8, CD45R, and CD11a (LFA-1, lymphocyte function-assocd. antigen 1) failed to evoke lytic activity. Thus, CD44 and gp90MEL-14 mols., in addn. to participating in T-cell homing and adhesion, may play a major role in **delivering** the transmembrane signal to the CTL that triggers the lytic activity, even when the T-cell **receptor** is not occupied. Such a mechanism may account for the nonspecific tissue damage seen at sites of CTL-mediated inflammation.

L22 ANSWER 8 OF 8 HCAPLUS COPYRIGHT 1996 ACS
AN 1986:606946 HCAPLUS
DN 105:206946
TI Targeted inhibition of transferrin-mediated iron uptake in Hep G2 hepatoma cells
AU Wu, George Y.; Wu, Catherine H.
CS Sch. Med., Univ. Connecticut, Farmington, CT, 06032, USA
SO J. Biol. Chem. (1986), 261(36), 16834-7
CODEN: JBCHA3; ISSN: 0021-9258
DT Journal
LA English
AB A model system consisting of 2 human hepatoma cell lines, Hep G2, representing well differentiated, normal hepatocytes, and PLC/PRF/5, representing poorly differentiated malignant hepatocytes, was used to demonstrate that the differential presence of asialoglycoprotein **receptor** activity in these cell lines can be used to influence transferrin-mediated Fe uptake. When added to culture medium, 55Fe-transferrin complexes **delivered** 55Fe well to both cell lines. As expected, in the presence of 55Fe-transferrin complexes, free primaquine caused a concn.-dependent decrease in 55Fe uptake in both cell lines. To create a targetable conjugate, primaquine was covalently coupled to asialofetuin to form asialofetuin-primaquine. When PLC/PRF/5 (asialoglycoprotein **receptor-neg**.) cells were preincubated with this conjugate, transferrin-mediated 55Fe uptake was unaffected. However, transferrin-mediated 55Fe uptake by Hep G2 (asialoglycoprotein **receptor-pos**.) cells under identical conditions was specifically decreased by 55% compared to control cells incubated without the conjugate.

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=> D HIS

     (FILE 'HCAPLUS' ENTERED AT 07:20:14 ON 25 SEP 96)
                DEL HIS   Y
L1         48268 S DELIVER?
L2           622 S L1 AND (POS OR POSITIV? OR POLYCATION? OR CATION?) AND
L3            17 S L2 AND (POLYMAINE# OR SPERMIDINE? OR POLYLYSINE OR POLY
L4            52 S L2 AND (POLYNUCLEO? OR DNA OR NUCLEIC OR OLIGONUCLEO?)
L5             5 S L2 AND POLYAMINE?
L6            19 S L3 OR L5
L7             9 S L6 AND L4
L8            21 S L2 AND CELL####(3A)TARGET?
L9             6 S L8 AND POLYMER?
L10           13 S L7 OR L9
L11            7 S L8 AND (L3 OR L4 OR L5)
L12           13 S L7 OR L9 OR L11
L13            7 S L4 AND CELL####(3A)TARGET?
L14            1 S L13 AND (CONTRAST? OR IMAG?)
L15          855 S  L1 AND CELL####(3A)TARGET?
L16          263 S RECEPTOR? AND L15
L17           12 S L16 AND L2
L18            8 S L2 AND CELL####(4A)UPTAKE?
L19            0 S L18 AND HYDROPHOB?
L20            0 S L14 NOT L12
L21            0 S L13 NOT L12
L22            8 S L17 NOT L12
L23            6 S L18 NOT (L12 OR L17)

=> D 1-6 BIB ABS

L23  ANSWER 1 OF 6  HCAPLUS  COPYRIGHT 1996 ACS
AN   1996:470845  HCAPLUS
DN   125:150920
TI   Receptor-mediated cell specific delivery of drugs to the
     liver and kidney
AU   Hashida, Mitsuru; Nishikawa, Makiya; Takakura, Yoshinobu
CS   Faculty Pharmaceutical Sciences, Kyoto University, Kyoto, 606-01,
     Japan
SO   Adv. Biomater. Biomed. Eng. Drug Delivery Syst., [Iketani Conf.
     Biomed. Polym.], 5th (1996), Meeting Date 1995, 86-90.  Editor(s):
     Ogata, Naoya. Publisher: Springer, Tokyo, Japan.
     CODEN: 63CXA6
DT   Conference
LA   English
AB   Effectiveness of several approaches aiming at hepatic and renal
     targeting of drugs and proteins is compared from various aspects.
     Receptor-mediated endocytosis of macromols. with sugar moieties,
     scavenger receptor-mediated endocytosis of polyanions, and
     general electrostatic interaction of polycations with cell
     surfaces are characterized through pharmacokinetic anal. at a whole
     body level in order to evaluate their potentials in drug targeting.
     Based on the obtained results, superoxide dismutase (SOD) was
     derivatized to various forms and mannosylated SOD and
   cationized SOD showed inhibitory effect against injury
     induced by ischemia/reperfusion in the liver and kidney, resp.  Mol.
     design of a carrier system with galactose residue was further
     discussed and it was concluded that the cellular
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uptake rate of macromols. was controlled by the d. of galactose on the mol. surface. The maximal affinity was given at surface d. of higher than 1.0 .times. 10-3 mols./.ANG.2 in the case of globular proteins. However, higher extent of modification is required in the case of vitamin K5 conjugate utilizing poly(L-glutamic acid) (PLGA) as a carrier backbone.

L23 ANSWER 2 OF 6 HCAPLUS COPYRIGHT 1996 ACS
AN 1995:753046 HCAPLUS
DN 123:237695
TI **Cell uptake** of albumin with synthetic glycolipids
AU Sato, Toshinori; JsooMiyaguchi, Hajime; Okahata, Yoshio
CS Dep. Biomol. Eng., Tokyo Inst. Technol., Yokoyama, 226, Japan
SO Drug Delivery Syst. (1995), 10(3), 199-200
CODEN: DDSYEI; ISSN: 0913-5006
DT Journal
LA Japanese
AB Internalization of albumin with synthetic glycolipid into blood cell was investigated by flow-cytometer. 1,5-Dioctyl-N-glucono-L-glutamate (2C8-glc) and dodecylmaltoside (DDM) enhanced the uptake of albumin into monocytes, but not into neutrophiles and granulocytes. **Cationic** lipids and **anionic** lipids showed no enhancement of **cell uptake** of albumin. Hemolysis was occurred in the presence of DDM, but not in the presence of 2C8-glc. The present study suggest that synthetic glycolipid 2C8-glc is expected as a candidate for the **delivery** of proteins into blood monocytes.

L23 ANSWER 3 OF 6 HCAPLUS COPYRIGHT 1996 ACS
AN 1995:265364 HCAPLUS
DN 122:47331
TI Physiological significance of IGF-I and its binding proteins on fetal growth and maturation
AU Iwashita, Mitsutoshi
CS Tokyo Women's Medical College Maternal and Perinatal, Tokyo, Japan
SO Nippon Sanka Fujinka Gakkai Zasshi (1994), 46(8), 660-72
CODEN: NISFAY; ISSN: 0300-9165
DT Journal
LA Japanese
AB The physiol. significance of IGF-I and IGF-BPs on fetal growth was studied in women. In the mother, circulating levels of IGF-I increased during pregnancy, whereas IGF-BPs, except IGF-BP-1, decreased. IGF-I stimulated [3H]-AIB uptake and release by cultured trophoblast cells in a dose-dependent manner. Furthermore, fetal growth and transfer of [3H]AIB to the fetus was inhibited when IGF-I was neutralized by polyclonal antibodies. These results indicate that maternal IGF-I stimulates fetal growth by activating placental transport of nutrients to the fetus. In contrast, IGF-BP-1 inhibited both 125I-IGF-I binding to placental membrane and IGF-I stimulation of [3H]glycine **uptake** by trophoblast **cells** in a dose-dependent manner. Moreover, fetal growth and the transfer of [3H]AIB to the fetus were accelerated when IGF-BP-1 was neutralized by polyclonal antibodies, suggesting that maternal IGF-BP-1 inhibits fetal growth by inhibiting IGF-I action on the placenta. IGF-I and IGF-BP-1, -2, -3, and -4 were localized in cytotrophoblast of term placenta. Similarly, IGF-BP-1, -2, and -4 were diminished by IGF-I and all 3 IGF-BPs were increased by

progesterone. Thus, there is a complicated autocrine/paracrine regulation between decidua and placenta and IGF-I action on fetal growth. Fetal levels of IGF-I and IGF-BP-1 were **pos**. and **neg**. correlated with fetal wt., resp. The isomers of phosphorylated IGF-BP-1 in cord sera were sepd. by **anion** ion exchange chromatog., and 1 nonphosphorylated and 4 phosphorylated IGF-BP-1 forms were detected. In paired blood samples from mid-term **delivery**, the percentage of nonphosphorylated IGF-BP-1 was higher in fetal blood than in maternal blood. Similarly, the percentage of nonphosphorylated IGF-BP-1 was higher in AFD infants than in SFD infants from term **delivery**. Thus, the proportion of nonphosphorylated and phosphorylated isomers of IGF-BP-1 varied in relation to fetal growth. Nonphosphorylated IGF-BP-1 enhanced IGF-I-mediated [3H]AIB **uptake** by cultured fibroblast **cells**, whereas phosphorylated IGF-BP-1 inhibited it. Affinity crosslinking expts. demonstrated that downregulation of receptors for IGF-I was prevented when cells were incubated with IGF-I in the presence of nonphosphorylated IGF-BP-1, whereas cell surface receptors were markedly reduced by downregulation of receptors when cells were incubated with IGRF-I alone or with phosporylated IGF-BP-1. Furthermore, IGF-I-induced desensitization of these cells in terms of [3H]AIB **uptake** was inhibited when **cells** were incubated with IGF-I in the presence of nonphosphorylated IGF-BP-1. Thus, the mechanism by which nonphosphorylated IGF-BP-1 potentiates IGF-I action was elucidated, and this mechanism may explain the remarkable fetal growth. The no. and vol. of lamellar bodies in lung of fetal mice increased when IGF-I was neutralized while lung maturity was inhibited after neutralization of IGF-I, suggesting that fetal IGF-I and IGF-BP-1 are involved not only in fetal growth but also in maturation. IGF-I is only one of may factors that influence fetal growth; however, it is notable that this growth factor is directly involved in fetal growth and maturation by modifying feto-maternal interaction.

L23 ANSWER 4 OF 6 HCAPLUS COPYRIGHT 1996 ACS
AN 1990:96630 HCAPLUS
DN 112:96630
TI Transfer of preformed terminal C5b-9 complement complexes into the outer membrane of viable gram-**negative** bacteria: effect on viability and integrity
AU Tomlinson, Stephen; Taylor, Peter W.; Luzio, J. Paul
CS Dep. Clin. Biochem., Univ. Cambridge, Cambridge, CB2 2QR, UK
SO Biochemistry (1990), 29(7), 1852-60
CODEN: BICHAW; ISSN: 0006-2960
DT Journal
LA English
OS CJACS
AB An efficient fusion system between gram-**neg**. bacteria and liposomes incorporating detergent-extd. complement C5b-9 complexes has been developed that allows **delivery** of preformed terminal complexes to the cell envelope (S. Tomlinson et al., 1989b). Fusion of Salmonella minnesota Re595 and Escherichia coli 17 with C5b-9-incorporated liposomes resulted in the transfer of 1900 C5b-9 complexes to each target bacterial cell. No loss in viability of bacteria was obsd. following fusion, even though the deposition of 900 complexes onto the envelope following exposure to lysozyme-free serum effected a >99% loss of viability. Increased

sensitivity to antibiotics normally excluded from the cell by an integral outer membrane (OM), as well as the ability of the chromogenic substrate PADAC to gain access to periplasmically located .beta.-lactamase, indicated that transferred C5b-9 complexes functioned as water-filled channels through the OM. A similar conclusion was drawn from measurements demonstrating the **uptake** by **cells** of the lipophilic **cation** tetraphenylphosphonium bromide, further indicating that the membrane potential across the cytoplasmic membrane was maintained following C5b-9 transfer to the OM. Examn. of S. minnesota Re595 by electron microscopy revealed no obvious difference between cells exposed to lethal concns. of lysozyme-free serum and cells following fusion with C5b-9-incorporated liposomes. These data suggest either that there are crit. sites in the OM to which liposome-**delivered** C5b-9 complexes are unable to gain access or that bacterial cell death is related to events occurring during polymn. of C9 on the cell surface.

L23 ANSWER 5 OF 6 HCAPLUS COPYRIGHT 1996 ACS
AN 1988:110616 HCAPLUS
DN 108:110616
TI Enhanced antiproliferative action of interferon targeted by bispecific monoclonal antibodies
AU Alkan, Sefik S.; Towbin, Harry; Hochkeppel, Heinz Kurt
CS Pharm. Res. Div., Ciba-Geigy Ltd., Basel, CH-4002, Switz.
SO J. Interferon Res. (1988), 8(1), 25-33
CODEN: JIREDJ; ISSN: 0197-8357
DT Journal
LA English
AB It has previously been shown that interferon (IFN) can be coupled covalently to tumor-specific monoclonal antibodies (mAb) and that the in vitro antiviral and antiproliferative action of these IFN-mAb conjugates is superior to that of uncoupled IFN. Here, a different mode of IFN **delivery** is reported, i.e., via bispecific mAbs, avoiding chem. coupling of IFN. Bispecific mAbs were prepd. by cross-linking 2 mAbs with SPDP, mAb1 being specific for an idiotype of a hybridoma cell-surface Ig and mAb2 specific for an IFN. Alternatively, Fab' fractions of mAb1 and mAb2 were coupled by disulfide formation to produce F(ab')2. Binding capacity and specificity of both arms of the mAb conjugates were first demonstrated by a solid-phase RIA using idiotype-**pos**. mAb as test antigen and 125I-labeled hybrid IFN-.alpha.B/D. Secondly, hybridomas either idiotype **pos**. or **neg**. were incubated with bispecific mAbs (mAb1-mAb2 or Fab'1-Fab'2) and 125I-labeled IFN at 4.degree.. After washing away unbound reagents, the **uptake** of radioactivity into **cells** was detd. Addnl., the antiproliferative action of cold or labeled IFN targeted via different modes was assessed by an [3H]TdR incorporation method. The bispecific mAbs specifically **delivered** IFN to the target cells and also inhibited their growth in vitro. Furthermore, targeting IFN by any of the 3 methods, IFN-mAb, mAb1-mAb2, or Fab'1-Fab'2, enhanced its in vitro antiproliferative potency compared to IFN alone.

L23 ANSWER 6 OF 6 HCAPLUS COPYRIGHT 1996 ACS
AN 1982:622846 HCAPLUS
DN 97:222846
TI Uptake of liposomes and liposome-encapsulated muramyl dipeptide by

human peripheral blood monocytes

AU Mehta, K.; Lopez-Berestein, G.; Hersh, E. M.; Juliano, R. L.
CS Med. Sch., Univ. Texas, Houston, TX, 77025, USA

SO J. Reticuloendothel. Soc. (1982), 32(2), 155-64
CODEN: JRSODF; ISSN: 0033-6890
DT Journal
LA English

AB The interaction of multilamellar liposomes with human peripheral blood monocytes, cultured in vitro was examd. Phagocytic engulfment is the principal mechanism by which the liposomes are taken up by these cells. Studies with radiolabeled liposomes revealed that they are taken up by monocytes as intact structures. The uptake is temp. sensitive and is affected by inhibitors of glycolysis and of microfilament activity. Monocytes take up **neg**. charged vesicles more rapidly than **pos**. charged (3-fold) vesicles or neutral vesicles (5-fold). Increase in **neg**. charge of liposomes enhances their **uptake** by the **cells**, but increased satn. of the phospholipids results in decreased uptake. Liposomes provide an effective means of enhancing the uptake of muramyl dipeptide (MDP) [53678-77-6] derivs. (3H nor MDP) by monocytes, with a 20-fold greater uptake of liposome encapsulated drug than of the free compd. Monocytes do not degrade the 3H nor MDP that they have internalized, and the radiolabel is only slowly released from the cells. These observations suggest a pharmacokinetic basis for the use of lipid vesicles as a system for the **delivery** of immunomodulating drugs to monocytes.

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=> D HIS L24-

     (FILE 'HCAPLUS' ENTERED AT 07:20:14 ON 25 SEP 96)
                SAV JONES/L ALL

     FILE 'WPIDS' ENTERED AT 08:24:28 ON 25 SEP 96
L24             8 S L12
L25             1 S L14
L26             1 S L17
L27             6 S L18

=> S L24-L27
L28          10 (L24 OR L25 OR L26 OR L27)

=> D 1-10 BIB ABS

L28  ANSWER 1 OF 10  WPIDS   COPYRIGHT 1996 DERWENT INFORMATION LTD
AN   96-342230 [34]   WPIDS
DNC  C96-108719
TI   New expanded porphyrin analogue turcasarin cpds. - used as metal
     chelating agents, radiation sensitisers for tumour therapy and
     transporting agents for e.g. antiviral drugs.
DC   B02
IN   BRUCKER, E A; SESSLER, J L; WEGHORN, S J
PA   (TEXA) UNIV TEXAS SYSTEM
CYC  60
PI   WO 9621665  A1 960718 (9634)* EN  104 pp
        RW: AT BE CH DE DK ES FR GB GR IE IT KE LU MC MW NL OA PT SD SE
            SZ
         W: AM AT AU BB BG BR BY CA CH CN CZ DE DK EE ES FI GB GE HU JP
            KE KG KP KR KZ LK LR LT LU LV MD MG MN MW MX NL NO NZ PL PT
            RO RU SD SE SI SK TJ TT UA US UZ VN
ADT  WO 9621665 A1 WO 95-US530 950113
PRAI WO 95-US530    950113
AN   96-342230 [34]   WPIDS
AB   WO 9621665 A   UPAB: 960829
     Decapyrrolic expanded porphyrins of formula (I), designated
     'turcasarins' and their salts are new. R1-R3 = H, alkyl (opt.
     substd. by halogen, OH, alkoxy, COOH, amino, sulphonato, ester,
     amide, phosphate, phosphonate or sulphate), alkenyl, alkynyl, opt.
     substd. aryl, amino, OH, alkoxy, carboxy, carboxamide, opt. substd.
     ester, opt. substd. amide, sulphonato, glycol, polyglycol, substd.
     ether, SH, alkylthiol, alkoxycarbonyl, aryloxycarbonyl, aldehyde,
     ketone, carboxylic acid (sic), phosphate, sulphate or phosphonate; a
     combination of the above; or gp. of formula -(CH2)mA(CH2)nB; A =
     CH2, O, S, NH, NR4, a divalent analogue of R1-R3, oxy, sulphide,
     thiol-substd. carboxamide or CONR4; R4 = alkyl, haloalkyl,
     hydroxyalkyl, glycol, polyglycol or alkyl thiol; B = nucleobase,
     saccharide, nucleotide, expanded porphyrin, steroid, aminoacid,
     peptide, polypeptide, turcasarin or polymeric or solid
     support matrix; m, n = 0-10; total C number in each of R1-R4 is not
     more than 20 (pref. at most 10).
          Also new are metal chelates (I') of (I), specifically where the
     metal is uranium.
          USE - (I) are metal chelating agents (claimed), and (I) and
     (I') are radiation sensitisers (claimed). They are useful in the
     photodynamic therapy of tumours, where they localise in tumours,
```

produce singlet oxygen and absorb at visible light wavelengths (700-800 nm) where body tissues are transparent.

As complexing agents they chelate, e.g., zinc or uranyl **cations**, and are useful, e.g., in removing uranium **cations** from solns.

(I) are also useful as drug carriers, since they can bind **anions** at near-neutral pH and transport **anions** across lipophilic structures such as biological membranes. Typically they are used for **delivery** of nucleobase antimetabolites (having antiviral, anticellular, antitumour, antiproliferative or antienzymatic activity) across biological membranes to facilitate **uptake** by **target cells**, esp. for treatment of viral diseases such as AIDS, herpes, hepatitis and measles; or in gene therapy for **delivery** of **oligonucleotides** or **DNA** fragments, e.g., for inhibiting an aberrant gene using an antisense **DNA** construct.

(I) are useful as chloride ion transporters for facilitating out-of-cell diffusion of chloride **anions**, esp. in the treatment of cystic fibrosis.

Dwg.0/7

L28 ANSWER 2 OF 10 WPIDS COPYRIGHT 1996 DERWENT INFORMATION LTD
AN 96-221768 [22] WPIDS
DNC C96-070320
TI Vehicle for **delivering nucleic** acid or therapeutic agents to cells - in complex of oppositely charged **polymers** including **cell targetting** gp. and with physiological agent attached, useful e.g. in gene therapy of cancer and for **imaging**.
DC A96 B07 D16
IN FRASER, S E; KAYYEM, J F; MEADE, T J
PA (CALY) CALIFORNIA INST OF TECHNOLOGY
CYC 66
PI WO 9611712 A2 960425 (9622)* EN 37 pp
RW: AT BE CH DE DK ES FR GB GR IE IT KE LU MC MW NL OA PT SD SE SZ UG
W: AL AM AT AU BB BG BR BY CA CH CN CZ DE DK EE ES FI GB GE HU IS JP KE KG KP KR KZ LK LR LT LU LV MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK TJ TM TT UA UG US UZ VN
AU 9641535 A 960506 (9636)
ADT WO 9611712 A2 WO 95-US14621 951011; AU 9641535 A AU 96-41535 951011
FDT AU 9641535 A Based on WO 9611712
PRAI US 94-321552 941012
AN 96-221768 [22] WPIDS
AB WO 9611712 A UPAB: 960604
Delivery vehicle comprises:(1) a first **polymer** (I) with a net **negative** or **positive** charge;(2) 1 second **polymer** (II) with a net charge opposite to that of (I) and complexed with it, (II) having a **cell targeting** gp. (III);(3) 1 physiological agent (IV) attached to (I) or (II), or to a third **polymer** (V), having a net charge opposite to that of (I) and complexed to it.

Also new are vehicles in which (I) has a net **positive** charge and which includes hydrophobic residues to facilitate **cellular uptake**, complexed to (II) of net **negative** charge, with (IV) attached to 1 component.

USE - The vehicles can be used to **deliver**:(a)

nucleic acid (NA), including antisense sequences, esp. for gene therapy of cancers, or(b) (IV), which are partic. **contrast** agents for magnetic resonance **imaging** (MRI) or anticancer agents (claimed). The vehicles are usually administered by injection but may also be **delivered** to the lungs as an aerosol.

ADVANTAGE - The vehicles bind specifically to, and are taken into, **target cells** and large numbers of (IV) can be attached without loss of this specificity, where (IV) includes an **imaging** agent, it allows **delivery** to the cell to be monitored (also the death of tumour cells can be detected). Dwg.2/4

L28 ANSWER 3 OF 10 WPIDS COPYRIGHT 1996 DERWENT INFORMATION LTD
AN 96-171606 [17] WPIDS
DNC C96-054191
TI **Delivery** of **nucleic** acids to cells, e.g. for gene therapy - using ternary complex of the **nucleic** acid with hydrophobic, membrane-binding **anion**, pref. hypericin, and a **polycation**, pref. **poly lysine**.
DC B04 D16
IN LAVIE, G; PRINCE, A M
PA (NYBL-N) NEW YORK BLOOD CENTER INC; (UYNY) UNIV NEW YORK STATE
CYC 19
PI WO 9607731 A1 960314 (9617)* EN 48 pp
RW: AT BE CH DE DK ES FR GB GR IE IT LU MC NL PT SE
W: AU CA JP
AU 9535894 A 960327 (9627)
ADT WO 9607731 A1 WO 95-US11709 950905; AU 9535894 A AU 95-35894 950905
FDT AU 9535894 A Based on WO 9607731
PRAI US 94-300725 940902
AN 96-171606 [17] WPIDS
AB WO 9607731 A UPAB: 960428
A new method for **delivering nucleic** acids to cells comprises: (i) forming a ternary complex between (a) a hydrophobic, membrane-binding **anion** selected from polycyclic aromatic diones of formula (I), anthraquinones of formula (II), emodin anthrone derivs. of formula (III), cercosporine derivs. of specified formulae, and fatty acids of formula CH3(CH2)nCOOH (V), or their salts; (b) a **polycation**; and (c) the **nucleic** acid; and (ii) contacting the cells with the ternary complex. In the formulae, R1 and R2 = H, OH, CH3, (CH2)nCH3, COCH3, CO(CH2)nCH3, CH(CH3)2 or COO(CH2)nCH3; n = 1 to 20; R3 and R4 = H, OH or COCH3; R1' = H, OH, CH3, COCH3, (CH2)nCH3 or CO(CH2)nCH3; R2' = H, OH or COCH3; and R4' = H, OH, COCH3, (CH2)nCH3 or CO(CH2)nCH3.

USE - The method is useful for transfection of cultured mammalian cells. It can be used to create protein 'factories' capable of producing large amounts of exogenous protein. The method can also be used in gene therapy, when it is desired to administer specific **nucleic** acids that encode a desired product to ameliorate a pathological condition.

ADVANTAGE - The complex facilitates the **delivery** and **uptake** of **DNA** into **cells**, specifically to overcome the hydrophobic barrier. Dwg.0/4

L28 ANSWER 4 OF 10 WPIDS COPYRIGHT 1996 DERWENT INFORMATION LTD

AN 95-328105 [42] WPIDS
DNC C95-145543
TI New cyclic conjugate of **polycationic polymer** and oligo nucleotide(s) - covalently bonded at each end by crosslinking agent, useful for anti sense and anti gene therapy, have strong binding to target and good in-vo stability.
DC A96 B04 D16
IN STEIN, S; TUNG, C; WEI, Z; ZHU, T
PA (UYNE-N) UNIV NEW JERSEY
CYC 51
PI WO 9524222 A1 950914 (9542)* EN 48 pp
RW: AT BE CH DE DK ES FR GB GR IE IT KE LU MC MW NL OA PT SD SE SZ UG
W: AM AT AU BB BG BR BY CA CH CN CZ DE DK ES FI GB GE HU JP KE KG KP MG MN MW NL NO NZ PL PT RO RU SD SE SI SK TJ TT UA UZ VN
AU 9521169 A 950925 (9601)
ADT WO 9524222 A1 WO 95-US2894 950307; AU 9521169 A AU 95-21169 950307
FDT AU 9521169 A Based on WO 9524222
PRAI US 94-207438 940307
AN 95-328105 [42] WPIDS
AB WO 9524222 A UPAB: 951026
New cyclic conjugate (A) comprises a **polycationic polymer** (PCP) covalently bonded at each end to 3' and 5' terminal nucleotides of a **polyanionic oligonucleotide** (ON) via a crosslinking agent. The PCP is of formula (I)-(VIII), in which X = NH, O or S; R1 = opt. substd. 1-4C alkyl; R2 and R3 = crosslinking agents covalently linking PCP and ON; R4 = opt. substd. 1-5C alkyl; R5 = prim., sec. or tert. amino, quat. ammonio, imidazole or guanidino; R6 = 1-2C alkyl residue; R7 = H, 1-5C alkyl, benzyl or (CH2)zCOR3; z = 0-3; a = 3-16; b, d, g = 2-5; c, e, h = 0-3; f = 2-9; i = 3-12; and the ratio of **cations** in PCP to **anions** in ON is 0.7-1.5:1.
USE - (A) are useful in antisense and antigene therapy.
ADVANTAGE - PCP improves binding of ON to complementary sequences (probably by restricting conformation and reducing ionic repulsion between ON sequences; PCP may also catalyse degradation of target RNA). (A) have high in vivo stability (since termini of ON are blocked); enter cells more easily than charged ON; and can be derivatised with intercalators, **cell targeting** agents, **delivery** systems etc. to improve activity. They are of low toxicity since degradation leads to nucleotides and amino acids only.
Dwg.0/6

L28 ANSWER 5 OF 10 WPIDS COPYRIGHT 1996 DERWENT INFORMATION LTD
AN 95-246111 [32] WPIDS
DNC C95-112911
TI Highly packed, poly **cationic** ammonium, sulphonium or phosphonium lipid(s) - useful for making lipid aggregates for **delivery** of, e.g., macromolecules into cells.
DC A26 A96 B04 B05 D16
IN CICCARONE, V C; HACES, A
PA (LIFE-N) LIFE TECHNOLOGIES INC
CYC 19
PI WO 9517373 A1 950629 (9532)* EN 50 pp
RW: AT BE CH DE DK ES FR GB GR IE IT LU MC NL PT SE
W: AU CA JP

AU 9514372 A 950710 (9543)
ADT WO 9517373 A1 WO 94-US14475 941214; AU 9514372 A AU 95-14372 941214
FDT AU 9514372 A Based on WO 9517373
PRAI US 93-171232 931220
AN 95-246111 [32] WPIDS
AB WO 9517373 A UPAB: 950818
Cpds. of formulae (I), (II) and (III): X = N, S, P or SO; x' = 1-20; ni (where i is 1 - x) = 1-6; l, m, n = 1-6; x = 1-10, Ra, Rb = H, 1-6C alkyl, 1-6C hydroxyalkyl, or 1-6C alkyl (substd. by thiol); R1-R4 = 1-6C alkyl; r = 0 or 1 (when X = N), 0 (when X = S or SO), or 1 (when X = P); A1-A4 = a gp. Z1-Z6; Z1 = a straight chain alkyl, alkenyl or alkynyl gp. contg. 2-22C in which 1 non-neighbouring CH2 gps. are opt. replaced by O or S; Z2 = a branched alkyl, alkenyl or alkynyl gp. contg. 2-22C in which 1 non-neighbouring CH2 gps. are opt. replaced by O or S; Z3 = a straight or branched chain alkyl gp. substd. by 1-2 OH, SH, NH2 or amine (sic) within 3C atoms of the bond between Z3 and X; Z4 = Z1 or Z2 (substd. by an aromatic, alicyclic, heterocyclic or polycyclic ring); Z5 = a gp. B-L; Z6 = a gp. CH(D-L)2 or C(D-L)3; B = CO, CO2, OCO, CON, OCON, OCH2, CH2O, SCH2, CH2S or CH2; D = CO, CO2, OCO, CON, OCON, O or S; L = Z1, Z2 or Z4, or an aromatic, alicyclic, heterocyclic or polycyclic moiety.

Cpds. of formula (IV), (V) and (VI) are also claimed; s = 2-6; R = R5-R8; R' = unbranched alkyl, alkenyl, alkynyl or alkoxy contg. 2-22C; Ar1, Ar2 = aryl rings; R5 = unbranched alkyl, alkenyl, alkynyl, or alkoxy contg. 2-22C; R6 = branched alkyl, alkenyl, alkynyl or alkoxy contg. 2-22C; R7 = an opt. substd. aromatic, dicyclic, heterocyclic or polycyclic gp; R8 = alkyl, alkenyl, alkynyl or alkoxy (each contg. 2-22C, and each substd. by an opt. substd. aromatic, alicyclic, heterocyclic or polycyclic ring).

Lipid aggregates comprising a cpd. (I)-(VI) are also claimed.

USE - Cpds. (I)-(VI) (cpds. (IV)-(VI) being cpds. (I)-(III)) are highly packed **polycationic** ammonium, sulphonium and phosphonium lipids and may be formed into liposomes or other lipid aggregates. These aggregates are **polycationic** and are able to form stable complexes with **anionic** macromolecules such as **nucleic** acids.

These **polyanion**-lipid complexes interact with cells making the **polyanionic** macromolecule available for absorption and **uptake** by the **cell**.

The complexes may be used for in vitro and in vivo transfection of cells (esp. eukaryotic cells), to generate transfected cells which generate useful gene prods. They may be used in cancer treatment, in in vivo and ex vivo gene therapy, and in diagnostic methods.

ADVANTAGE - The cpds. show unusually high affinity for the lipid bilayer of cell membranes as they comprise lipidic substits. at each **cationic** binding region.

They promote proximity between a complexed **polyanion** and a **target cell** membrane, thus increasing interactions between the two entities.

Dwg.0/0

L28 ANSWER 6 OF 10 WPIDS COPYRIGHT 1996 DERWENT INFORMATION LTD
AN 94-101064 [12] WPIDS
DNC C94-046559
TI New di hydroxypropyl alkylene di- and poly- amine **cationic** lipids - for conjugation with reporter molecules, **DNA**, or drugs, for assay, enhanced **targetting**, and transfection of

cells..
DC B04 B05 D16
IN CHYTIL, A; CICCARONE, V C; GEBEYEHU, G; HAWLEY-NELSON, P; JESSEE, J A
PA (LIFE-N) LIFE TECHNOLOGIES INC
CYC 20
PI WO 9405624 A1 940317 (9412)* EN 44 pp
RW: AT BE CH DE DK ES FR GB GR IE IT LU MC NL PT SE
W: CA JP
US 5334761 A 940802 (9430) 11 pp
EP 656883 A1 950614 (9528) EN
R: AT BE CH DE DK ES FR GB GR IE IT LI LU MC NL PT SE
EP 656883 A4 950809 (9618)
ADT WO 9405624 A1 WO 93-US8130 930827; US 5334761 A US 92-937508 920828; EP 656883 A1 EP 93-920406 930827, WO 93-US8130 930827; EP 656883 A4 EP 93-920406
FDT EP 656883 A1 Based on WO 9405624
PRAI US 92-937508 920828
AN 94-101064 [12] WPIDS
AB WO 9405624 A UPAB: 960705
2,3-Dihydroxypropyl alkylene di- and poly- amine derivs. and haloamine cpds. of formula (I) are new. In (I), R1, R2 = 1-23C alkyl or alkenyl, or 2-24C alkanoyl or alkenoyl; Z1, Z2 = H or 1-6 n-alkyl; q = 1-6; X = (CH2Onhalo, (CH2)nNH2, NH(CH2)mNH2, NH(CH2)3NH(CH2)4NH2, NH(CH2)3NH(CH2)4NH(CH2)3NH2, NHCOCH(NH(CH2)3NH2)(CH2)3NH(CH2)3NH2, NHCOCH(NH2)-(CH2)3NH2, NHCOCH(NHY)(CH2)pNH2, (CH2)rSH, or (CH2)sSS(CH2)tNH2; Y = H; n, r, s = 0-6; p = 2-5; and m, t = 2-6.
USE/ADVANTAGE - (I) are **cationic** lipids, and the halo cpds. are intermediates. The **cationic** cpds. are useful either alone, or in combination with lipid aggregating cpds., for formulation into liposomes or other lipid aggregates. These are able to complex with **anionic** macromolecules, including **nucleic** acids. The complex is then able to interact with cells to make the macromolecule available for absorption and **uptake** by the **cell**, e.g., in a transfection process. The halo cpds., and other (I) if linked through a crosslinking agent, can react with reporter molecules (e.g., fluorophors, luminophors, dyes, biotin), proteins, polypeptides, antibodies, **polyamines**, polyamino acids and polysaccharides, to permit targeted **delivery** or its assessment. They can also conjugate with solid support materials for use, e.g., in sepns. They can also react with **DNA** intercalating cpds., drugs, other therapeutic cpds., to enhance **delivery** efficiency. (I) are improvements on the known DOTMA, DOPE, DOGS and DPPES. They have enhanced applicability to different types of cells and **delivery** cpds., are stable and less toxic to **target cells**.
Dwg.1/4
Dwg.1/4
ABEQ US 5334761 A UPAB: 940914
Cationic lipids of formula
R1OCH2CH(OR2)CH2N+(Z1)(Z2)(CH2)qX1-16 (I)
are new (where R1 and R2 are each 1-23C alkyl or alkenyl or CO 1-23C alkyl or alkenyl; Z1 and Z2 are each H or 1-6C linear alkyl; q is 1-6; X is X1-X8, X15 or X16; X1 is (CH2)nQ; Q is F, Br, Cl or I; n is 0-6; X2 is (CH2)nNH2; X3 is NH(CH2)mNH2; m is 2-6; X4 is NH(CH2)3-NH-(CH2)4NH2; X5 is NH(CH2)3NH(CH2)4NH(CH2)3NH2; X6 is

NH-CO-CH(NH(CH2)3NH2)-(CH2)-3-NH(CH2)3NH2; X7 is NHCOCH(NH2)-(CH2)3NH2; X8 is NHCOCH(NHY)(CH2)pNH2; X15 is (CH2)rSH; X16 is (CH2)5S-S-(CH2)t-NH2; s is 0-6; t is 2-6; and r is 0-6).

USE - For making lipid aggregates for **delivery** of macromolecular and other cpds. into cells. They are esp. used for **DNA** development transformation of cells.

Dwg.0/0

L28 ANSWER 7 OF 10 WPIDS COPYRIGHT 1996 DERWENT INFORMATION LTD
AN 92-150593 [18] WPIDS
DNC C92-069689
TI Modification of target tissue with ion pair conjugates - e.g. pairs using tetra choro-platinate and organic-**cations**, for treatment of e.g. leukaemia.
DC B02
IN RIDEOUT, D C
PA (SCRI) SCRIPPS RES INST
CYC 15
PI WO 9205803 A 920416 (9218)* EN 17 pp
RW: AT BE CH DE DK ES FR GB GR IT LU NL SE
W: CA JP
EP 551442 A1 930721 (9329) EN 17 pp
R: DE FR GB
JP 06504773 W 940602 (9426) 6 pp
EP 551442 A4 950802 (9617)
ADT WO 9205803 A WO 91-US7292 911004; EP 551442 A1 WO 91-US7292 911004, EP 92-901703 911004; JP 06504773 W WO 91-US7292 911004, JP 92-502214 911004; EP 551442 A4 EP 92-901703
FDT EP 551442 A1 Based on WO 9205803; JP 06504773 W Based on WO 9205803
PRAI US 90-592926 901004
AN 92-150593 [18] WPIDS
AB WO 9205803 A UPAB: 931006
A **target** organism, **cell** or tissue is modified by exposing it to a **positive** ion component (I) and a **negative** ion component (II) which undergo ion-pairing in the microenvironment of the target to form an active ion-pair bonded conjugate (A). Also new are compsns. contg. (I) and (II).

Both components are pref. supplied as water-soluble salts. (I) is specifically dequalinium or rhodamine-123 ion while (II) is tetrachloroplatinate. Opt. at least one component coupled to a targer-specific ligand (**receptor** ligand, antibody or immunologically reactive fragment).

USE/ADVANTAGE - Use of (I) and (II) as separate soluble components facilitates **delivery** of (A) to target sites, specifically tumours and infectious agents, both in vivo and in vitro. Formation of the ion-pair may reduce toxicity of the individual components, permitting higher doses to be admin. (0/4) 0/4

ABEQ EP 551442 A UPAB: 931116
A **target** organism, **cell** or tissue is modified by exposing it to a **positive** ion component (I) and a **negative** ion component (II) which undergo ion-pairing in the microenvironment of the target to form an active ion-pair bonded conjugate (A). Also new are compsns. contg. (I) and (II).

Both components are pref. supplied as water-soluble salts. (I) is specifically dequalinium or rhodamine-123 ion while (II) is tetrachloroplatinate. Opt. at least one component coupled to a target-specific ligand (**receptor** ligand, antibody or

immunologically reactive fragment).
USE/ADVANTAGE - Use of (I) and (II) as separate soluble components facilitates **delivery** of (A) to target sites, specifically tumours and infectious agents, both in vivo and in vitro. Formation of the ion-pair may reduce toxicity of the individual components, permitting higher doses to be admin..

L28 ANSWER 8 OF 10 WPIDS COPYRIGHT 1996 DERWENT INFORMATION LTD
AN 91-339519 [46] WPIDS
DNN N91-260165 DNC C91-146521
TI New **cationic** lipid cpds. - used to enhance **delivery** of biologically active agents into cells of plants and animals.
DC B04 B05 B07 C03 D16 D21 P32
IN BASAVA, C; BORDER, R C; FELGNER, P L; HWANG-FELGNER, J; KUMAR, R; HWANGFELGN, J Y
PA (VICA-N) VICAL INC
CYC 17
PI WO 9116024 A 911031 (9146)*
RW: AT BE CH DE DK ES FR GB GR IT LU NL SE
W: AU JP
AU 9178547 A 911111 (9207)
EP 523189 A1 930120 (9303) EN 97 pp
R: AT BE CH DE DK ES FR GB GR IT LI LU NL SE
US 5264618 A 931123 (9348) 41 pp
JP 05508626 W 931202 (9402) 34 pp
US 5459127 A 951017 (9547) 44 pp
EP 523189 A4 950426 (9614)
ADT EP 523189 A1 EP 91-908905 910418, WO 91-US2691 910418; US 5264618 A Cont of US 90-511219 900419, Cont of US 90-563444 900807, US 91-686746 910416; JP 05508626 W JP 91-508835 910418, WO 91-US2691 910418; US 5459127 A Cont of US 90-511219 900419, Div ex US 90-563444 900807, Div ex US 91-686746 910416, US 93-123757 930916; EP 523189 A4 EP 91-908905
FDT EP 523189 A1 Based on WO 9116024; JP 05508626 W Based on WO 9116024; US 5459127 A Div ex US 5264618
PRAI US 91-686746 910416; US 90-511219 900419; US 90-563444 900807; US 93-123757 930916
AN 91-339519 [46] WPIDS
AB WO 9116024 A UPAB: 930928
A **cationic** lipid of formula (I) is claimed. In (I), Y1, Y2= -O-CH2-, OC(O) or O; R1, R2=H or 1-23C alkyl or alkenyl; R3, R4=H or 1-24C alkyl; R5=1-24C alkyl; R6=C(O)-(CH2)m-NH, a diaminocarboxylic acid which is alkyl, aryl or aralkyl, or C(O)-(CH2)m-NH- linked to the diaminocarboxylic acid, or is absent; R7=H, spermine, **spermidine**, a histone or a protein with **DNA**-binding specificity, or the same gp. in which the amines of the R7 moiety are quarternised with R3, R4 or R5 gps.; or R7= an L- or D-alpha amino acid having a **positively** charged gp. on the side chain, the amino acids comprising arginine, histidine, lysine or ornithine or analogues or where the amine of the R7 moiety is quaternised with R3, R4 or R5 gps.; or R7 is a polypeptide selected from L- or D-alpha amino acids, where at least one of the amino acid residues is arginine, histidine, lysine, ornithine or analogues; n=1-8; m=1-18; X=a a non-toxic **cation**. @(97pp Dwg.No.0/19)@
ABEQ US 5264618 A UPAB: 940120
Cationic lipids of formula (I) are new.

Y1 and Y2 are -O-C(O)- or -O-; R1 is H or 1-24C alkyl or -alkenyl; R2 is 1-24C-alkyl or -alkenyl; R3 and R4 are 1-24C alkyl or H; R5 is 1-24C alkyl; R6 is -C(O)-(CH2)m-NH-, or alkyl, aryl or aralkyl diamino carboxylate ester gp. or both linked together; R7 is H, spermine, **spermidine**, a histone or protein with **DNA**-binding specificity opt. with the amine functionalities of R7 quaternised with R3-R5; or R7 is L- or D-alpha aminoacid with **positive** gp. on side chain, viz. Arg, His, Lys or Orn or analogs opt. with the amine of R7 quaternised with R3-R6; or R7 is a polypeptide contg. above aminoacid(s) or analogs; n is 1-8; m is 1-18; and X is non-toxic **anion**.

Specifically new cpds. include DL-1,2-dioleoyl-3-dimethylamino propyl-beta-hydroxyethyl ammonium or the salts.

USE - Facilitating the transport of bioactive agents into cells including transfection with **polynucleotides**, and parenteral or topical **delivery** of antivirals and immunogenic peptides.

Dwg.0/9

ABEQ US 5459127 A UPAB: 951128

Polynucleotides are transfected into cells using an effective transfection promoting amount of a lysophosphatide of formula (I) or (II), where Y = -O-CH2- or -O-C(O)-; R = 10-23C alkyl or alkenyl; Z = phospholipid head gp. A formulation for transfecting **polynucleotides** and peptides into cells is esp. a cpd. of formula (III) or its optical isomer together with an effective transfecting amount of a lysophosphatide, esp. phosphatidylcholine or -ethanolamine. In (III), Y' = -O- or -O-C(O)-; R' = H, 1-14C alkyl, 7-11C aryl or alkaryl; 2 R''' together can form quinuclidino, piperidino, pyrrolidino or morpholino; X = non-toxic **anion** . The **cationic** lipid is e.g. DOTMA, DOTAP, esp. in the form of vesicles in an aq. medium. The formulation also contains a) a neutral lipid, e.g. cholesterol, b) a lysolipid, e.g. lysophosphatidylcholine and c) a therapeutic agent, e.g. a corticosteroid.

USE/ADVANTAGE - To enhance **delivery** of biologically active agents, esp. **polynucleotides**, peptides, proteins or drug molecules by facilitating transmembrane transport or encouraging adhesion to biological surfaces. Stable or transient transfection into cells is carried out more effectively than previously. The **cationic** lipids are metabolisable to reduce their in vivo and in vitro toxicity.

Dwg.0/0

L28 ANSWER 9 OF 10 WPIDS COPYRIGHT 1996 DERWENT INFORMATION LTD
AN 91-294949 [40] WPIDS
CR 86-184601 [29]; 90-090410 [12]; 90-260352 [34]; 93-159116 [19]; 95-005780 [01]; 96-383620 [38]
DNN N91-225937 DNC C91-127515
TI Use of quat. ammonium di ether(s) in liposome(s) - for transdermal, topical or ocular drug **delivery**.
DC B05 B07 C02 C03 D16 P32
IN EPPSTEIN, D A; FELGNER, P L; GADEK, T R; JONES, G H; ROMAN, R B
PA (SYNT) SYNTEX (USA) INC
CYC 1
PI US 5049386 A 910917 (9140)*
ADT US 5049386 A US 90-524257 900515
PRAI US 90-524257 900515; US 85-689407 850107; US 86-877916 860624; US 87-114809 871029; US 89-428815 891027

AN 91-294949 [40] WPIDS
CR 86-184601 [29]; 90-090410 [12]; 90-260352 [34]; 93-159116 [19]; 95-005780 [01]; 96-383620 [38]
AB US 5049386 A UPAB: 960924
A method for transdermal, topical or ocular **delivery** of a drug to the skin or to a mucous membrane of a human or animal subject, comprise: (i) forming a liposome comprising a drug and a dialkoxy-or dialkenyl- alkyltrialkylammonium lipid of formula (I) or an optical isomer, and (ii) applying the liposome to the skin or mucous membrane. Where R1, R2 = 6-24C alkyl, R3, R4, R5 = 1-8C alkyl, 6-11C aryl, or 7-11C aralkyl, or two of R3-R5 together = pyrrolidino, piperidino or morpholino, the third = 1-8C alkyl, 6-11C aryl or 7-11C aralkyl, or all three, R3-R5 = quinuclidino, X = an **anion** and n = 1-8.

USE/ADVANTAGE - (I) are **cationic** lipophilic cpds. and are used for the formulation of **positively** charged liposomes. The drug incorporated can be hydrophobic or hydrophilic. The (I) permit up to 100% entrapment of **polyanionic** substances for **delivery** in convenient manner. One such **polyanion** is DNA, which has e.g. an antigen, can be transfected into cells. The presentation of **positively** charged material to the **negatively** charged **cell** membrane results in better **uptake** of biological material by the cell. The liposomes using (I) have a geometry more compatible with the formation of bilayers, leading to increased physical stability. The ether linkage is stable, (I) are not leached out of the liposome matrix. @(33pp Dwg.No.0/1)@
0/1

L28 ANSWER 10 OF 10 WPIDS COPYRIGHT 1996 DERWENT INFORMATION LTD
AN 88-015589 [03] WPIDS
CR 91-150202 [21]
DNC C88-006722
TI PH sensitive immuno-conjugates - comprise antibody reactive with tumour associated antigens, therapeutic agent and linker for **delivery** of agent to tumour tissues.
DC B04
IN HELLSTROM, I E; HELLSTROM, K E; LAVIE, E
PA (BRIM) BRISTOL-MYERS SQUIBB CO; (ONCO) ONCOGEN; (BRIM) BRISTOL-MYERS CO
CYC 21
PI EP 253202 A 880120 (8803)* EN 34 pp
R: AT BE CH DE ES FR GB GR IT LI LU NL SE
AU 8774663 A 880107 (8810)
JP 63115827 A 880520 (8826)
US 4997913 A 910305 (9112)
AU 9178131 A 910829 (9141)
US 5084560 A 920128 (9207)
CA 1304293 C 920630 (9232)
AU 636493 B 930429 (9324)
JP 05088686 B 931224 (9403) 23 pp
ADT EP 253202 A EP 87-109414 870630; JP 63115827 A JP 87-162201 870629; US 4997913 A US 87-47161 870512; US 5084560 A US 90-564387 900807; CA 1304293 C CA 87-541057 870630; AU 636493 B AU 91-78131 910604, Div ex AU 87-74663 ; JP 05088686 B JP 87-162201 870629
FDT AU 636493 B Previous Publ. AU 9178131; JP 05088686 B Based on JP 63115827
PRAI US 86-880674 860630; US 87-47161 870512; US 86-880574 860630

AN 88-015589 [03] WPIDS
CR 91-150202 [21]
AB EP 253202 A UPAB: 940303
pH-sensitive immunoconjugate (I) for **delivering** a chemotherapeutic agent (II) to tumour tissues comprises (a) an antibody reactive with tumour associated antigens; (b) (II); and (c) a link between the antibody and (II), the link being unstable at low pH values. (I) dissociates in low pH tumour tissue and releases (I) in the tissue.

Pref. the pH unstable link is an amide bond and (I) has the structure of formula (III). X = amino gp. of (II); Y = amino gp. of a lysine residue of the antibody.

The antibody is isolated from polyclonal sera or is a monoclonal antibody, e.g. L6 and the antigen is L6 antigen. (II) is esp. daunomycin, mitomycin C, adriamycin or methotrexate. (I) releases (II) at pH 4-7, esp. 5.8-6.7. The spacer may be a polyaminoacid, esp. **poly**-L-**lysine** or HSA. Other linking gps. include diazo-benzyl bonds.

USE/ADVANTAGE - (I) are unstable at lower pH values, esp. when (II) is daunomycin, and they dissociate in human tumour tissues to **deliver** to (II) for a therapeutic effect. (I) may be radiolabelled so that on admin. the dose/g (I) required for a therapeutic effect can be determined.

Dwg.0/15

ABEQ US 4997913 A UPAB: 930923
A pH-sensitive immunoconjugate comprises (a) an antibody reactive with antigens of interest; (b) an anthrocycline chemotherapeutic agent with suitable toxicity; (c) link between (a) and (b) to form immunoconjugate, which comprises a spacer contg. three amino acids and is unstable in the pH range 5.1-7. The antibody is not internalised by **target** tumour **cells**. The immunoconjugate dissociates to release the chemotherapeutic agent outside **target cells** killing **positive** and **negative** antigen cells.

USE/ADVANTAGE - Used specifically for **delivering** chemotherapeutic agent to tumour cells. The pH sensitivity allows accurate targetting and release of the agent. @tig

ABEQ US 5084560 A UPAB: 930923
Prodn. of pH-sensitive immunoconjugates of an antibody to a tumour antigen and a chemotherapeutic agent comprises reaction of aconitic anhydride with a free amine gp. of the chemotherapeutic agent; condensn. of the resulting free COOH gp. with the terminal amine gp. of an aminoacid type of spacer; and then reaction with an activating agent to produce an active functional gp.; also, condensn. of a lysine gp. of the antibody with a thiolating agent; and then condensn. of the activated antibody deriv. with the chemotherapeutic cpd. deriv. Typical spacer is **poly**-L-**lysine**.

USE - The prods. have improved target-seeking properties, resulting in enhanced activity against tumours.

```
=> D HIS L29-

     (FILE 'MEDLINE' ENTERED AT 08:31:41 ON 25 SEP 96)
L29            9 S L12
L30            0 S L14
L31            9 S L17
L32            6 S L18
L33           21 S L29-L32
```

=> D L29 1-9 BIB ABS;D L31 1-9 BIB ABS;D L32 1-6 BIB ABS

L29 ANSWER 1 OF 9 MEDLINE
AN 96215256 MEDLINE
TI Folate-targeted, **anionic** liposome-entrapped **polylysine**-condensed **DNA** for tumor cell-specific gene transfer.
AU Lee R J; Huang L
CS Department of Pharmacology, University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania 15261, USA.
NC CA59327 (NCI)
HL50256 (NHLBI)
DK44935 (NIDDK)
+
SO JOURNAL OF BIOLOGICAL CHEMISTRY, (1996 Apr 5) 271 (14) 8481-7.
Journal code: HIV. ISSN: 0021-9258.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals; Cancer Journals
EM 9608
AB We have developed a lipidic gene transfer vector, LPDII, where **DNA** was first complexed to **polylysine** at a ratio of 1:0.75 (w/w) and then entrapped into folate-targeted pH-sensitive **anionic** liposomes composed of dioleoyl phosphatidylethanolamine (DOPE)/cholesteryl hemisuccinate/folate-polyethlene glycol-DOPE (6:4:0.01 mol/mol) via charge interaction. LPDII transfection of KB cells, a cell line overexpressing the tumor marker folate receptor, was affected by both the lipid to **DNA** ratio and the lipid composition. At low lipid to **DNA** ratios (e.g. 4 and 6), LPDII particles were **positively** charged; transfection and cellular uptake levels were independent of the folate receptor and did not require a pH-sensitive lipid composition. Meanwhile, transfection and uptake of **negatively** charged LPDII particles, i.e. those with high lipid to **DNA** ratios (e.g. 10 and 12), were folate receptor-dependent and required a pH-sensitive lipid composition. The transfection activity of LPDII was lost when the inverted cone-shaped DOPE was replaced by dioleoyl phosphatidylcholine. LPDII particles with lipid to **DNA** ratios of 4, 6, 10, and 12 were approximately 20-30 times more active than **DNA** .3-beta-[N-(N',N'-dimethylethane)carbamoyl]cholesterol **cationic** liposome complexes in KB cells and were much less cytotoxic. On the sucrose gradient, LPDII particles had a migration rate in between those of the free **DNA** and the **DNA** .**polylysine** complex. An electron micrograph of LPDII showed a structure of spherical particles with a **positively** stained core enclosed in a lipidic envelope with a mean diameter of 74 +/- 14 nm. This novel gene transfer vector may potentially be useful in gene therapy for tumor-specific **delivery**.

L29 ANSWER 2 OF 9 MEDLINE
AN 96146439 MEDLINE
TI Potentiation of **cationic** liposome-mediated gene **delivery** by **polycations**.
AU Gao X; Huang L
CS Department of Pharmacology, University of Pittsburgh School of

Medicine, Pennsylvania 15261, USA.
NC HL 50256 (NHLBI)
CA 59327 (NCI)
DK 44935 (NIDDK)
SO BIOCHEMISTRY, (1996 Jan 23) 35 (3) 1027-36.
Journal code: A0G. ISSN: 0006-2960.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 9604
AB We discovered that several high molecular weight **cationic** polymers, such as **poly**(L-**lysine**) and protamine, can enhance the transfection efficiency of several types of **cationic** liposomes by 2-28-fold in a number of cell lines in vitro. Small **polycations** such as spermine and a **cationic** decapeptide derived from SV40 T-antigen were only moderately active. The addition of **poly**(L-**lysine**) and protamine dramatically reduced the particle size of the complex formed between **DNA** and **cationic** liposomes and rendered **DNA** resistant to the nuclease activity. The complexes composed of **DNA**, **poly**(L-**lysine**), and **cationic** lipids were purified from an excess of free liposomes with sucrose gradient ultracentrifugation. Purified complex formed at low **cationic** liposome ratio was poor in lipid content and only had weak transfection activity. Addition of free liposome to the purified complex significantly enhanced the transfection activity. In contrast, complexes formed at a higher initial ratio of liposome to **DNA** had a higher lipid content and were highly active in transfection; the activity was about 3-9-fold more active than the corresponding complex before purification. **Negative** stain EM studies revealed that the most active complexes prepared from 40 nmol of lipid, 0.5 micrograms of **poly**(L-**lysine**), and 1 microgram of **DNA** and purified by gradient ultracentrifugation were spherical, electron dense, small (< 100 nm in diameter) particles, and some of them were associated with lipid membranes. These highly active, stable, small-sized lipid/**poly**(L-**lysine**)/**DNA** complexes represent a new class of nonviral gene **delivery** vehicles that might be useful in gene therapy.

L29 ANSWER 3 OF 9 MEDLINE
AN 96145365 MEDLINE
TI A total **delivery** system of genetically engineered drugs or cells for diseased vessels. Concept, materials, and fabricated prototype device.
AU Kito H; Suzuki T; Nagahara S; Nakayama Y; Tsutsui Y; Isutsui N; Nakajima N; Matsuda T
CS Department of Bioengineering, National Cardiovascular Center Research Institute, Osaka, Japan.
SO ASAIO JOURNAL, (1994 Jul-Sep) 40 (3) M260-6.
Journal code: BBH. ISSN: 1058-2916.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 9605
AB The development of a percutaneous procedure using a catheterized

system for diseased vessels has been increasingly in demand in conjunction with gene therapy using genetically engineered drugs (antisense) and cells. The authors' strategic concept realizes revascularization at narrowed, diseased sites and **delivery** of drugs or cells into the diseased tissues or **targeted cells**. An inflatable, drug-releasing double balloon is installed at the tip of a catheter. The outer balloon, fabricated with micropores (diameters of 20 and 30 mm) by an excimer laser ablation technique, releases a viscous solution containing a photoreactive **polymer** and drug or cells on inflation of the inner balloon. A photoresponsive water-soluble **polymer**, molecularly designed for its ability to achieve prolonged local residency of antisense **DNA** at the tissue level and enhanced transmembrane transport at the cellular level, is premixed with antisense **oligonucleotide** drug. On light irradiation, the nonionic **polymer** is reversibly converted to a **positively** charged **polymer** that can be complexed with highly **negatively** charged antisense **DNA** (c-myb), which may enhance the transmembrane **delivery** of antisense. On cessation of irradiation, the complex slowly dissociates to function intracellularly as an antisense drug, resulting in inhibition of cell proliferation. Thus, our integrated, dual-function balloon system may contribute to mechanical dilatation gene therapies at diseased vessels.

L29 ANSWER 4 OF 9 MEDLINE
AN 95365355 MEDLINE
TI A versatile vector for gene and **oligonucleotide** transfer into cells in culture and in vivo: polyethylenimine.
AU Boussif O; Lezoualc'h F; Zanta M A; Mergny M D; Scherman D; Demeneix B; Behr J P
CS Laboratoire de Chimie Genetique, Centre National de la Recherche Scientifique, Faculte de Pharmacie, Illkirch, France..
SO PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1995 Aug 1) 92 (16) 7297-301.
Journal code: PV3. ISSN: 0027-8424.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals; Cancer Journals
EM 9511
AB Several **polycations** possessing substantial buffering capacity below physiological pH, such as lipopolyamines and polyamidoamine **polymers**, are efficient transfection agents per se--i.e., without the addition of **cell targeting** or membrane-disruption agents. This observation led us to test the **cationic polymer** polyethylenimine (PEI) for its gene-**delivery** potential. Indeed, every third atom of PEI is a protonable amino nitrogen atom, which makes the **polymeric** network an effective "proton sponge" at virtually any pH. Luciferase reporter gene transfer with this **polycation** into a variety of cell lines and primary cells gave results comparable to, or even better than, lipopolyamines. Cytotoxicity was low and seen only at concentrations well above those required for optimal transfection. **Delivery** of **oligonucleotides** into embryonic neurons was followed by using a fluorescent probe. Virtually all neurons showed nuclear labeling, with no toxic effects. The optimal PEI **cation**/

anion balance for in vitro transfection is only slightly on the **cationic** side, which is advantageous for in vivo **delivery**. Indeed, intracerebral luciferase gene transfer into newborn mice gave results comparable (for a given amount of **DNA**) to the in vitro transfection of primary rat brain endothelial cells or chicken embryonic neurons. Together, these properties make PEI a promising vector for gene therapy and an outstanding core for the design of more sophisticated devices. Our hypothesis is that its efficiency relies on extensive lysosome buffering that protects **DNA** from nuclease degradation, and consequent lysosomal swelling and rupture that provide an escape mechanism for the PEI/**DNA** particles.

L29 ANSWER 5 OF 9 MEDLINE
AN 95330815 MEDLINE
TI Natural killer (NK) activity in human responders and nonresponders to stimulation by anti-CD16 antibodies.
AU Galatiuc C; Gherman M; Metes D; Sulica A; DeLeo A; Whiteside T L; Herberman R B
CS Center for Immunology, Bucharest, Romania..
SO CELLULAR IMMUNOLOGY, (1995 Jul) 163 (2) 167-77.
Journal code: CQ9. ISSN: 0008-8749.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals; Cancer Journals
EM 9510
AB Various anti-Fc gamma RIII (CD16) monoclonal antibodies (mAbs) are shown here to have **positive** or **negative** modulatory effects on human NK cells. Thus, 3G8 mAb (IgG1) triggered a dose-dependent augmentation of NK activity in 67% (23/34) of individuals tested, who were designated as responders. All four IgG1 anti-CD16 mAb tested (BL-LGL/1, B73.1, Leu11c, and 3G8) were stimulatory for NK cells isolated from responders, whereas six non-IgG1 anti-CD16 mAbs were either inhibitory or had no significant effects on NK activity. The upregulation of NK activity in responders was not attributable to an increase in either the conjugate formation or the **delivery** of the lethal hit to **target cells**. This mAb-mediated up-regulation of NK activity was shown to be associated with a recycling capacity higher than that of controls and with enhanced release of cytokines by activated NK cells. Anti-CD16 mAb inhibited binding of either monomeric or **polymeric** IgG to Fc gamma RIIIA on NK cells. Also, mAb 3G8 or its F(ab')2 fragments decreased or reversed inhibition of NK activity induced by monomeric IgG (mIgG). Our data indicate that regulation of NK activity via the Fc gamma RIIIA is influenced by dose-dependent interactions between cytophilic mIgG and anti-CD16 mAb of IgG1 isotype.

L29 ANSWER 6 OF 9 MEDLINE
AN 95053755 MEDLINE
TI **Delivery** of a viral antigen to the class I processing and presentation pathway by Listeria monocytogenes.
AU Ikonomidis G; Paterson Y; Kos F J; Portnoy D A
CS Department of Microbiology, University of Pennsylvania School of Medicine, Philadelphia 19104-6076..
NC AI-27655 (NIAID)
GM-31841 (NIGMS)

SO JOURNAL OF EXPERIMENTAL MEDICINE, (1994 Dec 1) 180 (6) 2209-18. Journal code: I2V. ISSN: 0022-1007.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals; Cancer Journals
EM 9502
AB Listeria monocytogenes is a facultative intracellular pathogen that grows in the cytoplasm of infected host cells. We examined the capacity of L. monocytogenes to introduce influenza nucleoprotein (NP) into the class I pathway of antigen presentation both in vitro and in vivo. Recombinant L. monocytogenes secreting a fusion of listeriolysin O and NP (LLO-NP) **targeted** infected **cells** for lysis by NP-specific class I-restricted cytotoxic T cells. Antigen presentation occurred in the context of three different class I haplotypes in vitro. A hemolysin-**negative** L. monocytogenes strain expressing LLO-NP was able to present in a class II-restricted manner. However, it failed to **target** infected **cells** for lysis by CD8+ T cells, indicating that hemolysin-dependent bacterial escape from the vacuole is necessary for class I presentation in vitro. Immunization of mice with a recombinant L. monocytogenes strain that stably expressed and secreted LLO-NP induced NP-specific CD8+ cytotoxic T lymphocytes. These studies have implications for the use of L. monocytogenes to **deliver** potentially any antigen to the class I pathway in vivo.

L29 ANSWER 7 OF 9 MEDLINE
AN 93195054 MEDLINE
TI Targeted transfection and expression of hepatitis B viral **DNA** in human hepatoma cells.
AU Liang T J; Makdisi W J; Sun S; Hasegawa K; Zhang Y; Wands J R; Wu C H; Wu G Y
CS Gastrointestinal Unit, Medical Services, Massachusetts General Hospital, Boston 02114..
NC DK-01952 (NIDDK)
CA-54524 (NCI)
CA-46801 (NCI)
+
SO JOURNAL OF CLINICAL INVESTIGATION, (1993 Mar) 91 (3) 1241-6. Journal code: HS7. ISSN: 0021-9738.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Abridged Index Medicus Journals; Priority Journals; Cancer Journals
EM 9306
AB A soluble **DNA** carrier system consisting of an asialoglycoprotein covalently linked to **poly**-L-**lysine** was used to bind **DNA** and **deliver** hepatitis B virus (HBV) **DNA** constructs to asialoglycoprotein receptor-**positive** human hepatoma cells. 4 d after transfection with surface or core gene expression constructs, HBsAg and HBeAg in the media were measured to be 16 ng/ml and 32 U/ml per 10(7) cells, respectively. Antigen production was completely inhibited by the addition of an excess of asialoorosomucoid. On the other hand, asialoglycoprotein receptor-**negative** human hepatoma cells, SK-Hep1, did not produce any viral antigens under identical conditions after incubation with HBV

DNA complexed to a conjugate composed of asialoorosomucoid and **poly**-L-**lysine**. Using a complete HBV genome construct, HBsAg and HBeAg levels reached 16 ng/ml and 16 U/ml per 10(7) cells, respectively. Northern blots revealed characteristic HBV RNA transcripts including 3.5-, 2.4-, and 2.1-kb fragments. Intracellular and extracellular HBV **DNA** sequences including relaxed circular, linear and single stranded forms were detected by Southern blot hybridization. Finally, 42-nm Dane particles purified from the spent cultures medium were visualized by electron microscopy. This study demonstrates that a targetable **DNA** carrier system can transfect HBV **DNA** in vitro resulting in the production of complete HBV virions.

L29 ANSWER 8 OF 9 MEDLINE
AN 92364367 MEDLINE
TI Class I restricted CTL recognition of a soluble protein **delivered** by liposomes containing lipophilic polylysines.
AU Nair S; Zhou X; Huang L; Rouse B T
CS Department of Microbiology, University of Tennessee, Knoxville 37996..
NC AI24762-05 (NIAID)
SO JOURNAL OF IMMUNOLOGICAL METHODS, (1992 Aug 10) 152 (2) 237-43. Journal code: IFE. ISSN: 0022-1759.
CY Netherlands
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals; Cancer Journals
EM 9211
AB CD8+ cytotoxic lymphocytes recognize peptides derived from endogenous antigens complexed with class I major histocompatibility complex while CD4+ helper cells recognize peptides from exogenous antigens bound to class II MHC molecules. A soluble protein can be introduced into the class I pathway of antigen processing and presentation using an appropriate vehicle to **deliver** the antigen into the cytosol. **Cationic** liposomes containing lipophilic **polylysine** readily form complexes with an **anionic**, soluble protein ovalbumin. Mouse thymoma EL4 cells incubated with such complexes can be sensitized for killing by OVA-specific CTL effector **cells**. This method of **target** sensitization by a soluble antigen is more sensitive than the osmotic loading method previously reported.

L29 ANSWER 9 OF 9 MEDLINE
AN 90234702 MEDLINE
TI Transfer of preformed terminal C5b-9 complement complexes into the outer membrane of viable gram-**negative** bacteria: effect on viability and integrity.
AU Tomlinson S; Taylor P W; Luzio J P
CS Department of Clinical Biochemistry, University of Cambridge, Addenbrooke's Hospital, U.K..
SO BIOCHEMISTRY, (1990 Feb 20) 29 (7) 1852-60. Journal code: A0G. ISSN: 0006-2960.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 9008
AB An efficient fusion system between Gram-**negative** bacteria

and liposomes incorporating detergent-extracted C5b-9 complexes has been developed that allows **delivery** of preformed terminal complexes to the cell envelope (Tomlinson et al., 1989b). Fusion of Salmonella minnesota Re595 and Escherichia coli 17 with C5b-9-incorporated liposomes resulted in the transfer of 1900 C5b-9 complexes to each **target** bacterial **cell**. No loss in viability of bacteria was observed following fusion, even though the deposotion of 900 complexes onto the envelope following exposure to lysozyme-free serum effected a greater than 99% loss of viability. Increased sensitivity to antibiotics normally excluded from the cell by an integral outer membrane (OM), as well as the ability of the chromogenic substrate PADAC to gain access to periplasmically located beta-lactamase, indicated that transferred C5b-9 complexes functioned as water-filled channels through the OM. A similar conclusion was drawn from measurements demonstrating the uptake by cells of the lipophilic **cation** tetraphenylphosphonium (bromide), a result further indicating that the membrane potential across the cytoplasmic membrane was maintained following C5b-9 transfer to the OM. Examination of S. minnesota Re595 by electron microscopy revealed no obvious difference between cells exposed to lethal concentrations of lysozyme-free serum and cells following fusion with C5b-9-incorporated liposomes. These data suggest either that there are critical sites in the OM to which liposome-**delivered** C5b-9 complexes are unable to gain access or that bacterial cell death is related to events occurring during **polymerization** of C9 on the cell surface.

L31 ANSWER 1 OF 9 MEDLINE
AN 95330815 MEDLINE
TI Natural killer (NK) activity in human responders and nonresponders to stimulation by anti-CD16 antibodies.
AU Galatiuc C; Gherman M; Metes D; Sulica A; DeLeo A; Whiteside T L; Herberman R B
CS Center for Immunology, Bucharest, Romania..
SO CELLULAR IMMUNOLOGY, (1995 Jul) 163 (2) 167-77.
Journal code: CQ9. ISSN: 0008-8749.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals; Cancer Journals
EM 9510
AB Various anti-Fc gamma RIII (CD16) monoclonal antibodies (mAbs) are shown here to have **positive** or **negative** modulatory effects on human NK cells. Thus, 3G8 mAb (IgG1) triggered a dose-dependent augmentation of NK activity in 67% (23/34) of individuals tested, who were designated as responders. All four IgG1 anti-CD16 mAb tested (BL-LGL/1, B73.1, Leu11c, and 3G8) were stimulatory for NK cells isolated from responders, whereas six non-IgG1 anti-CD16 mAbs were either inhibitory or had no significant effects on NK activity. The upregulation of NK activity in responders was not attributable to an increase in either the conjugate formation or the **delivery** of the lethal hit to **target cells**. This mAb-mediated up-regulation of NK activity was shown to be associated with a recycling capacity higher than that of controls and with enhanced release of cytokines

by activated NK cells. Anti-CD16 mAb inhibited binding of either monomeric or polymeric IgG to Fc gamma RIIIA on NK cells. Also, mAb 3G8 or its F(ab')2 fragments decreased or reversed inhibition of NK activity induced by monomeric IgG (mIgG). Our data indicate that regulation of NK activity via the Fc gamma RIIIA is influenced by dose-dependent interactions between cytophilic mIgG and anti-CD16 mAb of IgG1 isotype.

L31 ANSWER 2 OF 9 MEDLINE
AN 94354631 MEDLINE
TI Signal requirement for induction of MHC-unrestricted antitumor cytotoxicity of human T cell CD4+/CD8+ subpopulations.
AU Zhu H G; Klein-Franke A; Anderer F A
CS Friedrich-Miescher-Laboratorium der Max-Planck-Gesellschaft, Tuebingen, Germany..
SO ANTICANCER RESEARCH, (1994 May-Jun) 14 (3A) 953-61. Journal code: 59L. ISSN: 0250-7005.
CY Greece
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals; Cancer Journals
EM 9412
AB The role of cosignalling in the generation of MHC-unrestricted cytotoxicity of T cells was studied with CD4+ and CD8+ sub-populations highly purified (> 98%) by immunomagnetic cell sorting using OKT4 mab, Dynal anti-CD4 mab, OKT8 mab, Dynal anti-CD8 mab, and OKT3 mab. Cytotoxicity was determined in 4 h cytotoxicity assays against K562 tumor cells known to lack expression of MHC class 1 and class 2 antigens, thus avoiding interference with anti-CD4- or anti-CD8-mediated signalling. Signal transfer was induced via CD4, CD8, CD3, IL-2 **receptor** and RG **receptor** specifically interacting with a plant rhamnogalacturonan (RG). In CD8+ cells, the first signal **delivered** by the sorting mab (immobilized OKT8 or Dynal anti-CD8 or OKT3) only induced low MHC-unrestricted cytotoxicity but committed the cells to develop largely enhanced cytolytic potential upon stimulation with a second (IL-2 or RG) or third (OKT3, IL-2, RG) signal. The highest cytolytic potential was achieved by cumulative signalling via CD8, CD3, IL-2 **receptor** and RG **receptor**. The generation of MHC-unrestricted cytotoxicity of CD8+ cells correlated with increased effector **cell/target cell** conjugate formation. In CD4+ cells, OKT4 as sorting mab induced very low cytolytic potential, and a moderate committment to IL-2 signals but a stronger one to RG signals, yielding further cytotoxicity enhancement. The highest cytolytic potential was obtained by cumulative signalling via CD4, IL-2 **receptor** and RG **receptor**. Dynal anti-CD4 mab was inefficient and OKT3, as sorting mab of CD4+ cells from CD8-depleted PNAC, appeared to block subsequent OKT4-induced generation of MHC-unrestricted cytotoxicity by **delivering** a **negative** signal. Immobilized OKT3 as second signal present in cultures of OKT4-sorted CD4+ cells was inefficient. Surprisingly, soluble OKT3 together with IL-2 **delivered** a **positive** signal in cultures of OKT4-sorted CD4+ cells.

L31 ANSWER 3 OF 9 MEDLINE
AN 94340599 MEDLINE
TI Epidermal growth factor **receptors** in human breast

carcinoma **cells**: a potential selective **target** for transforming growth factor alpha-Pseudomonas exotoxin 40 fusion protein.
AU Arteaga C L; Hurd S D; Dugger T C; Winnier A R; Robertson J B
CS Department of Medicine, Vanderbilt University School of Medicine, Nashville, Tennessee 37232..
SO CANCER RESEARCH, (1994 Sep 1) 54 (17) 4703-9.
Journal code: CNF. ISSN: 0008-5472.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals; Cancer Journals
EM 9411
AB Epidermal growth factor (EGF) **receptors** are expressed in high levels by some poor prognosis breast tumors. We have examined the cytotoxic effect of the tumor growth factor alpha (TGF alpha)-delta Cys-Pseudomonas exotoxin (PE40) recombinant fusion protein on normal and tumorigenic human breast epithelial cells in vitro and in vivo. The MDA-468, MDA-231, BT-20, and MCF-7ADR estrogen **receptor-negative**, EGF **receptor** -rich breast cancer lines were exquisitely sensitive in vitro to TGF alpha-delta Cys-PE40 with a 50% inhibitory concentration of < or = 0.02 nM. The estrogen **receptor-positive**, low EGF **receptor** MCF-7, ZR75-1, and T47D cells were less sensitive to the fusion toxin with a 50% inhibitory concentration of > 0.2 nM. The nontumorigenic cell lines 184, 184A1, and 184B5 were relatively resistant to TGF alpha-delta Cys-PE40 despite exhibiting high levels of EGF **receptors**. Continuous i.p. administration of TGF alpha-delta Cys-PE40 via an osmotic minipump at a dose of 0.4 microgram/g/day over 7 days inhibited MDA-468, MA-231, and BT-20 but not MCF-7 tumor growth in female athymic mice. Host tissue toxicity was not observed with this dose of TGF alpha-delta Cys-PE40. Mixed MDA-468/MCF-7 tumors were established in nude mice after coinoculation of both cell types in estrogen-supplemented animals. EGF **receptor** immunohistochemistry and immunoblot procedures indicated that TGF alpha-PE40 eliminated the MDA-468 cells while sparing the adjacent MCF-7 cells. By immunoblot, EGF **receptors** were consistently more abundant in tumor tissue than in adjacent nontumor tissue from the same mastectomy specimen (n = 7). These data support the notion that EGF **receptors** can be selectively targeted in human breast cancer cells for the **delivery** of antitumor agents. Further clinical studies with TGF alpha-delta Cys-PE40 and other chimeric toxins using the same **cellular target** will address this possibility.

L31 ANSWER 4 OF 9 MEDLINE
AN 94041135 MEDLINE
TI Functional studies of adhesion molecules on CD4-CD8- double **negative** T cells of autoimmune MRL/Mp-lpr/mice.
AU Wang W
CS Institute of Immunological Science, Hokkaido University, Sapporo, Japan..
SO HOKKAIDO IGAKU ZASSHI. HOKKAIDO JOURNAL OF MEDICAL SCIENCE, (1993 Sep) 68 (5) 755-66.
Journal code: GA9. ISSN: 0367-6102.
CY Japan
DT Journal; Article; (JOURNAL ARTICLE)
LA Japanese

FS Priority Journals
EM 9402
AB MRL/Mp-lpr/lpr (MRL-lpr) mice have been used for a model of human systemic lupus erythematosus. This strain of mice homozygous for an autosomal recessive mutation, lpr (lymphoproliferation), develops massive lymphadenopathy with the expansion of CD4-CD8- (double **negative**; DN) T cells. Recently it was demonstrated that lpr mice have defects in the gene of Fas antigen which mediates apoptosis, indicating a possibility of defect in **negative** selection of autoreactive T cells in the thymus of lpr mice. However, the mechanisms that control the accumulation of DN T cells in lymph nodes, and the involvement of DN T cells in the clinical manifestation of disease, have not been well understood. In this study, the expression of various cell adhesion molecules on lymphocytes from MRL-lpr mice was examined. The strong expression of CD44 antigen as well as heat stable antigen (HSA) on abnormal DN T cells of lymph nodes was characteristic in MRL-lpr mice. Furthermore, the accumulation of DN T cells in lymph nodes might result from augmented binding of lymphocytes to endothelial cell surface of lymph nodes, possibly due to the failure of Mel-14 antigen shedding from DN T cell surface. In addition, it was found that monoclonal antibodies reactive with cell adhesion molecules such as CD44, Mel-14, CD45R and HSA expressed on DN T cells, could trigger the lytic activity of DN T cells and redirected DN T cell-mediated lysis of Fc-**receptor-positive** **target cells** (EL-4). In contrast to T cell **receptor** (TCR)-mediated cytotoxicity, this redirected cytotoxicity was not inhibited by anti-lymphocyte function associated antigen-1 (LFA-1) antibody. Thus, cell adhesion molecules may play a major role in **delivering** the transmembrane signal to DN T cells of MRL-lpr mice that trigger the lytic activity. It is likely that DN T cells of MRL-lpr mice induce tissue damages by the interaction with ligand on vascular endothelium or extracellular matrix in vivo.

L31 ANSWER 5 OF 9 MEDLINE
AN 92379477 MEDLINE
TI Human melanoma targeting with alpha-MSH-melphalan conjugate.
AU Ghanem G E; Libert A; Arnould R; Vercammen A; Lejeune F
CS L.O.C.E., Jules Bordet Institute, Universite Libre de Bruxelles, Belgium..
SO MELANOMA RESEARCH, (1991 Jun-Jul) 1 (2) 105-14.
Journal code: BJR. ISSN: 0960-8931.
CY ENGLAND: United Kingdom
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 9212
AB A conjugate made of alpha-MSH as a drug carrier and melphalan has been designed in order to **target** human melanoma **cells**. Iodination of the alpha-MSH moiety led to a relatively stable tracer which could be easily separated and analysed by reverse phase high pressure liquid chromatography. The conjugate was found to be unstable at neutral pH and a serious denaturation can take place at concentrations exceeding 100 micrograms/ml, especially in plasma. **Receptor**-mediated cytotoxicity has been shown by the use of cultured alpha-MSH **receptor positive/negative** cells as well

as in vivo B16 murine melanoma model. Body distribution and uptake of the labelled compound were unaltered as compared to those of labelled free hormone. alpha-MSH **receptor** recognition properties also remained unchanged with a better apparent affinity of the conjugate probably due to the alkylating activity of melphalan itself. Using human melanoma dendritic cells expressing more than 10,000 alpha-MSH binding sites per cell as an in vitro model, we were able to demonstrate higher cytotoxicities as compared to melphalan-treated cells. In contrast, melanoma cells with low receptivity did not show higher cytotoxicity. P388D1 mouse plasmocytoma cells lacking **receptors** were much more sensitive to melphalan than the conjugate. This phenomenon appeared to be related with the number of binding sites expressed at the time of the experiment as well as cell differentiation and the doubling time. Our findings strongly support the concept of a **receptor**-mediated cytotoxicity and may enable the in vivo melphalan **delivery** to target tissues to be increased, achieving an improvement of drug penetration inside melanoma cells.

L31 ANSWER 6 OF 9 MEDLINE
AN 91352094 MEDLINE
TI T-cell-**receptor**-independent activation of cytolytic activity of cytotoxic T lymphocytes mediated through CD44 and gp90MEL-14.
AU Seth A; Gote L; Nagarkatti M; Nagarkatti P S
CS Department of Biology, Virginia Polytechnic Institute and State University, Blacksburg 24061..
NC CA 45009 (NCI)
CA 45010 (NCI)
SO PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1991 Sep 1) 88 (17) 7877-81.
Journal code: PV3. ISSN: 0027-8424.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals; Cancer Journals
EM 9112
AB CD44 is a transmembrane glycoprotein found on a variety of cells including those of myeloid and lymphoid origin. CD44 is highly conserved among various species and is involved in the homing of lymphocytes and monocytes to lymph nodes, Peyer's patches, and sites of inflammation. In the present study, we demonstrate that monoclonal antibody (mAb) 9F3, directed against murine phagocytic glycoprotein 1 (CD44) expressed on cytotoxic T lymphocytes (CTLs), can trigger the lytic activity of CTLs and redirect CTL-mediated lysis to antigen-**negative** Fc **receptor**-**positive target cells**. Similar redirected lysis was also inducible using mAb MEL-14, directed against the lymphocyte homing **receptor** for endothelium (gp90MEL-14). The redirected lysis induced by mAbs 9F3 and MEL-14 was similar to that induced by mAbs against the alpha beta T-cell **receptor** or CD3. In contrast, mAbs directed against CD8, CD45R, and CD11a (LFA-1, lymphocyte function-associated antigen 1) failed to evoke lytic activity. The current study demonstrates that CD44 and gp90MEL-14 molecules, in addition to participating in T-cell homing and adhesion, may play a major role in **delivering** the transmembrane signal to the CTL that triggers the lytic activity, even when the T-cell **receptor** is not

occupied. Such a mechanism may account for the nonspecific tissue damage seen at sites of CTL-mediated inflammation.

L31 ANSWER 7 OF 9 MEDLINE
AN 89345318 MEDLINE
TI Covalent and noncovalent protein binding of drugs: implications for hepatic clearance, storage, and cell-specific drug **delivery**
.
AU Meijer D K; van der Sluijs P
CS Department of Pharmacology and Therapeutics, University Center of Pharmacy, University of Groningen, The Netherlands..
SO PHARMACEUTICAL RESEARCH, (1989 Feb) 6 (2) 105-18. Ref: 159
Journal code: PHS. ISSN: 0724-8741.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LA English
FS Priority Journals
EM 8911
AB This review deals with the mechanisms by which the liver disposes of drugs that are covalently or noncovalently associated with proteins. Many drugs bind to plasma proteins such as albumin (mainly **anionic** compounds) and alpha 1-acid glycoprotein (**cationic** compounds). Nevertheless, the liver is able to clear such drugs efficiently from the circulation because of intrahepatic dissociation of the drug-protein complex. This clearance may involve spontaneous dissociation because of progressive removal of the unbound drug during liver passage, a process that can be rate limiting in hepatic uptake. Alternatively, the porous endothelial lining of the hepatic sinusoids may allow extensive surface interactions of the drug-protein complexes with hepatocytes, leading to facilitation of drug dissociation. Binding to plasma proteins and intracellular proteins in the cytoplasm or cell organelles is an important factor determining the hepatic storage and elimination rate of drugs. Drugs noncovalently associated with glycosylated proteins, which can be endocytosed by various liver cells, are not coendocytosed with such proteins. However, covalently bound drugs can be internalized by **receptor**-mediated endocytosis, which permits specific **targeting** to hepatocytes, endothelial **cells**, Kupffer cells, and lipocytes by coupling to different glycoproteins that are recognized on the basis of their terminal sugar. The endocytosed drug-carrier complex is routed into endosomes and lysosomes, where the active drug is liberated by cleavage of acid-sensitive linkages or proteolytic degradation of peptide linkers. This concept has been applied to antineoplastic, antiparasitic, and antiviral drugs.

L31 ANSWER 8 OF 9 MEDLINE
AN 89089568 MEDLINE
TI Immunospecific targeting of cytosine arabinonucleoside-containing liposomes to the idiotype on the surface of a murine B-cell tumor in vitro and in vivo.
AU Bankert R B; Yokota S; Ghosh S K; Mayhew E; Jou Y H
CS Department of Molecular Immunology, Roswell Park Memorial Institute, Buffalo, New York 14263..
NC CA33462

CA25253
CA22786
+
SO CANCER RESEARCH, (1989 Jan 15) 49 (2) 301-8.
Journal code: CNF. ISSN: 0008-5472.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals; Cancer Journals
EM 8904
AB A new tumor model is described that is suitable for the evaluation of antibody-directed drug-**delivery** protocols and a modification in the procedure for covalently coupling antibody to the surface of drug-containing liposomes is presented. These immunospecific liposomes containing cytosine arabinonucleoside (Ara-C) have been tested in vitro and in vivo for their ability to kill a B-**cell** tumor. The **target** of the immunospecific-Ara-C liposomes is the idiotype associated with an antigen-specific immunoglobulin **receptor** on the cell surface of a murine B-cell hybrid (2C3). Affinity-purified antibodies specific for the idiotype were covalently coupled to modified lipid on the surface of the large unilamelar liposomes containing drug. These liposomes were shown to kill idiotype-**positive** 2C3 cells in vitro, but not idiotype-**negative** variants of this same cell line. It was also established in vitro that the drug-containing liposomes were at least 40 times more efficient than free Ara-C in the killing of the tumor cells. The 2C3 tumor was also propagated in vivo following the i.p. administration of tumor cells. The tumor grew initially as multiple foci within the peritoneum and subsequently spread to the spleen. Tumor-bearing mice were treated either with free Ara-C or with immunospecific liposomes containing Ara-C. Tumor growth in the primary tumor nodules and in the spleen was monitored by the administration of bromodeoxyuridine to the tumor-bearing animals followed by the immunofluorescent staining of cells with a monoclonal anti-bromodeoxyuridine antibody to estimate the proportion of cells in S phase. Our data from five out of seven animal experiments shows that the immunospecific-Ara-C liposomes, but not free drug, reduced tumor growth in the spleen. However, neither the liposomes containing drug nor the free drug were able to alter the growth of the primary tumor nodules growing in the peritoneal cavity. These results suggest that immunospecific-Ara-C containing liposomes may be useful in conjunction with other cytoreductive protocols in controlling tumor growth or preventing the spread of the tumor to other sites, but that immunospecific-Ara-C containing liposomes by themselves are not likely to eliminate an established tumor in vivo. We also demonstrate here that the administration of immunospecific-Ara-C containing liposomes in an animal having high levels of circulating tumor-associated antigen (i.e., IgG containing the idiotype) represents a potential clinically relevant hazard which must be considered when designing antibody-directed drug-**delivery** protocols.

L31 ANSWER 9 OF 9 MEDLINE
AN 81054719 MEDLINE
TI **Receptor**-mediated endocytosis of antibody-opsonized liposomes by tumor cells.

AU Leserman L D; Weinstein J N; Blumenthal R; Terry W D
SO PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1980 Jul) 77 (7) 4089-93.
Journal code: PV3. ISSN: 0027-8424.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 8103
AB Specific **receptor**-mediated **delivery** of the contents of small, sonicated liposomes was studied with three murine tumor cell types: an IgG Fc **receptor-negative** nonphagocytic line (EL4); an Fc **receptor-positive** phagocytic line (P388D1); and an Fc **receptor-positive** nonphagocytic line (P388). The liposomes (formed from phosphatidylcholines, cholesterol, and dinitrophenyl-substituted phosphatidylethanolamine) contained carboxyfluorescein as a fluorescent marker and methotrexate as a pharmacologic agent. Binding and internalization of the liposomes were observed by fluorescence microscopy and measured by flow microfluorometry. The hapten-derivatized lipid was used as a binding point on the liposome for the antibody-combining site of the immunoglobulin. In the presence of IgG anti-dinitrophenyl, but not F(ab')2 or IgA anti-dinitrophenyl, liposomes bound to the Fc **receptor**-bearing cells. The liposomes underwent endocytosis by the P388D1 cells and, to a lesser extent, by the P388 cells. As measured by depression of [3H]deoxyuridine incorporation, methotrexate in IgG-opsonized liposomes had a much greater pharmacologic effect on the P388D1 cells than did the same amount in unopsonized liposomes or in free solution. This observation indicates that an appropriately chosen drug, incorporated in liposomes, can exert its effect on a cytoplasmic **target** after endocytosis. P388 **cells** showed a moderate effect of the drug in liposomes. Neither P388 nor P388D1 cells bound or ingested unopsonized liposomes, and the Fc **receptor-negative** EL4 line neither bound nor ingested opsonized liposomes. The data demonstrate specific interaction of opsonized liposomes with the cells' IgG Fc **receptor**.

L32 ANSWER 1 OF 6 MEDLINE
AN 96215256 MEDLINE
TI Folate-targeted, **anionic** liposome-entrapped polylysine-condensed DNA for tumor cell-specific gene transfer.
AU Lee R J; Huang L
CS Department of Pharmacology, University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania 15261, USA.
NC CA59327 (NCI)
HL50256 (NHLBI)
DK44935 (NIDDK)
+
SO JOURNAL OF BIOLOGICAL CHEMISTRY, (1996 Apr 5) 271 (14) 8481-7.
Journal code: HIV. ISSN: 0021-9258.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals; Cancer Journals

EM 9608
AB We have developed a lipidic gene transfer vector, LPDII, where DNA was first complexed to polylysine at a ratio of 1:0.75 (w/w) and then entrapped into folate-targeted pH-sensitive **anionic** liposomes composed of dioleoyl phosphatidylethanolamine (DOPE)/cholesteryl hemisuccinate/folate-polyethlene glycol-DOPE (6:4:0.01 mol/mol) via charge interaction. LPDII transfection of KB cells, a cell line overexpressing the tumor marker folate receptor, was affected by both the lipid to DNA ratio and the lipid composition. At low lipid to DNA ratios (e.g. 4 and 6), LPDII particles were **positively** charged; transfection and **cellular uptake** levels were independent of the folate receptor and did not require a pH-sensitive lipid composition. Meanwhile, transfection and uptake of **negatively** charged LPDII particles, i.e. those with high lipid to DNA ratios (e.g. 10 and 12), were folate receptor-dependent and required a pH-sensitive lipid composition. The transfection activity of LPDII was lost when the inverted cone-shaped DOPE was replaced by dioleoyl phosphatidylcholine. LPDII particles with lipid to DNA ratios of 4, 6, 10, and 12 were approximately 20-30 times more active than DNA.3-beta-[N-(N',N'-dimethylethane)carbamoyl]cholesterol **cationic** liposome complexes in KB cells and were much less cytotoxic. On the sucrose gradient, LPDII particles had a migration rate in between those of the free DNA and the DNA.polylysine complex. An electron micrograph of LPDII showed a structure of spherical particles with a **positively** stained core enclosed in a lipidic envelope with a mean diameter of 74 +/- 14 nm. This novel gene transfer vector may potentially be useful in gene therapy for tumor-specific **delivery**.

L32 ANSWER 2 OF 6 MEDLINE
AN 94376010 MEDLINE
TI Physiological significance of IGF-I and its binding proteins on fetal growth and maturation.
AU Iwashita M
CS Tokyo Women's Medical College Maternal and Perinatal..
SO NIPPON SANKA FUJINKA GAKKAI ZASSHI. ACTA OBSTETRICA ET GYNAECOLOGICA JAPONICA, (1994 Aug) 46 (8) 660-72. Ref: 10
Journal code: INR. ISSN: 0300-9165.
CY Japan
DT Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LA Japanese
FS Priority Journals
EM 9412
AB Insulin-like growth factor-I (IGF-I) is one of growth factors that circulates bound to specific, high affinity binding proteins (IGFBPs). Physiological significance of IGF-I and IGFBPs on fetal growth is investigated in this study. In mother, circulating levels of IGF-I are increased during pregnancy in which placental hormones take the place of pituitary GH to regulate IGF-I during pregnancy and correlates with fetal birth weight. IGFBPs except IGFBP-1 in the maternal circulation are markedly reduced compared to those of non pregnant women due to increased activity of protease(s) while IGFBP-1 gradually increased throughout pregnancy and **negatively** correlates with fetal weight. IGF-I stimulated 3H-AIB uptake and release by cultured trophoblast cells in a dose

dependent manner. Furthermore, fetal growth and the transfer of 3H-AIB to fetus is inhibited when IGF-I is neutralized by polyclonal antibody. These results indicate that maternal IGF-I stimulates fetal growth by activating placental transport of nutrients to fetus. In contrast, IGFBP-1 inhibits both 125I-IGF-I binding to placental membrane and 3H-glycine **uptake** of trophoblast **cells** by IGF-I in a dose dependent manner. Moreover, fetal growth and the transfer of 3H-AIB to fetus are accelerated when IGFBP-1 is neutralized by polyclonal antibody, suggesting that maternal IGFBP-1 inhibits fetal growth by inhibiting IGF-I action on the placenta. IGF-I and four IGFBPs including IGFBP-1, -2, -3, and -4 are localized in cytotrophoblast of term placenta. Similarly IGFBP-1, -2, and -4 are detected in medium conditioned by term decidua cells by Western ligand blot in which release of IGFBP-1 and -4 are diminished by IGF-I and all three IGFBPs are increased by progesterone. Thus, there is a complicated autocrine/paracrine regulation between decidua and placenta and IGF-I action on fetal growth is presumed to be modified by this local regulation. Fetal levels of IGF-I and IGFBP-1 are **positively** and **negatively** correlate with fetal weight, respectively. The isomers of phosphorylated IGFBP-1 in cord sera are separated by **anion** ion exchange chromatography in which one nonphosphorylated and four phosphorylated IGFBP-1 are detected. In pared blood samples from mid-term **delivery**, percentage of nonphosphorylated IGFBP-1 is higher in fetal blood compared to those in mother. Similarly, percentage of nonphosphorylated IGFBP-1 is elevated in AFD infants than is SFD infants from term **delivery**. Thus, the proportion of nonphosphorylated and phosphorylated isomers of IGFBP-1 varies corresponding to fetal growth.(ABSTRACT TRUNCATED AT 400 WORDS)

L32 ANSWER 3 OF 6 MEDLINE
AN 90234702 MEDLINE
TI Transfer of preformed terminal C5b-9 complement complexes into the outer membrane of viable gram-**negative** bacteria: effect on viability and integrity.
AU Tomlinson S; Taylor P W; Luzio J P
CS Department of Clinical Biochemistry, University of Cambridge, Addenbrooke's Hospital, U.K..
SO BIOCHEMISTRY, (1990 Feb 20) 29 (7) 1852-60.
Journal code: A0G. ISSN: 0006-2960.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 9008
AB An efficient fusion system between Gram-**negative** bacteria and liposomes incorporating detergent-extracted C5b-9 complexes has been developed that allows **delivery** of preformed terminal complexes to the cell envelope (Tomlinson et al., 1989b). Fusion of Salmonella minnesota Re595 and Escherichia coli 17 with C5b-9-incorporated liposomes resulted in the transfer of 1900 C5b-9 complexes to each target bacterial cell. No loss in viability of bacteria was observed following fusion, even though the deposotion of 900 complexes onto the envelope following exposure to lysozyme-free serum effected a greater than 99% loss of viability. Increased sensitivity to antibiotics normally excluded from the cell by an integral outer membrane (OM), as well as the ability of the

chromogenic substrate PADAC to gain access to periplasmically located beta-lactamase, indicated that transferred C5b-9 complexes functioned as water-filled channels through the OM. A similar conclusion was drawn from measurements demonstrating the **uptake** by **cells** of the lipophilic **cation** tetraphenylphosphonium (bromide), a result further indicating that the membrane potential across the cytoplasmic membrane was maintained following C5b-9 transfer to the OM. Examination of S. minnesota Re595 by electron microscopy revealed no obvious difference between cells exposed to lethal concentrations of lysozyme-free serum and cells following fusion with C5b-9-incorporated liposomes. These data suggest either that there are critical sites in the OM to which liposome-**delivered** C5b-9 complexes are unable to gain access or that bacterial cell death is related to events occurring during polymerization of C9 on the cell surface.

L32 ANSWER 4 OF 6 MEDLINE
AN 88214116 MEDLINE
TI Enhanced antiproliferative action of interferon targeted by bispecific monoclonal antibodies.
AU Alkan S S; Towbin H; Hochkeppel H K
CS Pharmaceuticals Research Division, Ciba-Geigy Limited, Basel, Switzerland..
SO JOURNAL OF INTERFERON RESEARCH, (1988 Feb) 8 (1) 25-33. Journal code: IJI. ISSN: 0197-8357.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 8808
AB It has previously been shown that interferon (IFN) can be coupled covalently to tumor-specific monoclonal antibodies (mAb) and that the in vitro antiviral and antiproliferative action of these IFN-mAb conjugates is superior to that of uncoupled IFN. We now report a different mode of IFN **delivery**, i.e., via bispecific mAbs, avoiding chemical coupling of IFN. Bispecific mAbs were prepared by cross-linking two mAbs with SPDP, mAb1 being specific for an idiotype of a hybridoma cell-surface immunoglobulin and mAb2 specific for an IFN. Alternatively, Fab' fractions of mAb1 and mAb2 were coupled by disulfide formation to produce F(ab')2. Binding capacity and specificity of both arms of the mAb conjugates were first demonstrated by a solid-phase radioimmunoassay using idiotype-**positive** mAb as test antigen and 125I-labeled hybrid IFN-alpha B/D. Secondly, hybridomas either idiotype **positive** or **negative** were incubated with bispecific mAbs (mAb1-mAb2 or Fab'1-Fab'2) and 125I-labeled IFN at 4 degrees C. After washing away unbound reagents, the **uptake** of radioactivity into **cells** was determined. Additionally, the antiproliferative action of cold or labeled IFN targeted via different modes was assessed by an [3H]TdR incorporation method. Results showed that bispecific mAbs could specifically **deliver** IFN to the target cells and also inhibit their growth in vitro. Furthermore, targeting IFN by any of the three methods, IFN-mAb, mAb1-mAb2, or Fab'1-Fab'2, enhanced its in vitro antiproliferative potency compared to IFN alone.

L32 ANSWER 5 OF 6 MEDLINE

AN 86274545 MEDLINE
TI In vitro genotoxicity studies using complex hydrophobic mixtures: efficient **delivery** of a petroleum sample to cultured C3H/10T1/2 cells via lipid vesicle incorporation.
AU von Hofe E H; Billings P C; Heidelberger C; Landolph J R
NC 3341-01
SO ENVIRONMENTAL MUTAGENESIS, (1986) 8 (4) 589-609. Journal code: EIY. ISSN: 0192-2521.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals; Cancer Journals
EM 8611
AB Petroleum fractions are a diverse group of extremely hydrophobic mixtures, some of which display strong carcinogenicity in animal skin painting experiments. Interpretation of in vitro genotoxicity experiments with these samples is complicated by inefficient **delivery** of these hydrophobic substances inside target cells. We therefore developed methods to assess and improve the efficiency of **delivering** a petroleum sample (Matrix, A.P.I. 81-17) to cultured C3H/10T1/2 cells for genotoxicity studies via lipid vesicle incorporation. Three radiolabeled compounds (14C-benzo(a)pyrene, 14C-decane, and 14C-naphthalene) of widely differing volatilities, broadly representative of the spectrum of compounds in petroleum samples, were separately added to Matrix. Lipid vesicles containing Matrix and radiolabeled compounds were prepared by the classical methods for preparing neutral, **positive**, and **negatively** charged multilamellar and unilamellar liposomes. Of these, the classical methods for preparing neutral unilamellar liposomes were the most successful for **delivering** radiolabeled compounds in Matrix to cells. Vesicles optimal for the **delivery** of tracers in Matrix were prepared with DSPC:cholesterol:lyso-PC (8.8:0.8:0.4, molar ratio) in a Matrix to lipid ratio of 31:69 (w/w). This new method of **delivery** resulted in proportional, dose-dependent, and reproducible **uptake** of all tracers. Further, **cells** treated with this preparation took up 2.5-fold more 14C-decane, 1.5-fold more 14C-BaP, and 18-fold more 14C-naphthalene added to Matrix than did cells treated with Matrix emulsified in tissue culture medium. In contrast, tracers were not taken up in a proportional or reproducible manner when emulsions were used, and in fact, uptake of 14C-naphthalene was consistently very small. Two petroleum fractions, C(2)029188 and C(3)029194, were 4- and 6-fold more cytotoxic, respectively, when **delivered** to C3H/10T1/2 cells by lipid vesicles than emulsions. The carcinogenic petroleum fraction C(5)0292202 induces type II transformed foci in C3H/10T1/2 cells when cells were treated with C(5)0292202 incorporated into lipid vesicles. The methods for lipid vesicle incorporation described here are effective in **delivering** hydrophobic petroleum fractions to cells and provide an alternative to the current inefficient and artifactual methods of emulsification currently used. With further validation and standardization, lipid vesicle incorporation of petroleum fractions and treatment of cells with these vesicles should be useful for studying the genotoxicity of complex hydrophobic mixtures in cell culture systems.

L32 ANSWER 6 OF 6 MEDLINE
AN 83138525 MEDLINE

TI Uptake of liposomes and liposome-encapsulated muramyl dipeptide by human peripheral blood monocytes.
AU Mehta K; Lopez-Berestein G; Hersh E M; Juliano R L
NC RR5511-9
CA 25129
SO JOURNAL OF THE RETICULOENDOTHELIAL SOCIETY, (1982 Aug) 32 (2) 155-64.
Journal code: JWV. ISSN: 0033-6890.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 8306
AB The interaction of multilamellar liposomes with human peripheral blood monocytes, cultured in vitro, has been examined. Phagocytic engulfment is the principal mechanism by which the liposomes are taken up by these cells. Studies with radiolabeled liposomes revealed that they are taken up by monocytes as intact structures. The uptake is temperature sensitive and is affected by inhibitors of glycolysis and of microfilament activity. Monocytes take up **negatively** charged vesicles more rapidly than **positively** charged (3-fold) vesicles or neutral vesicles (5-fold). Increase in **negative** charge of liposomes enhances their **uptake** by the **cells**, but increased saturation of the phospholipids results in decreased uptake. Liposomes provide an effective means of enhancing the uptake of MDP derivatives (3H nor MDP) by monocytes, with a 20-fold greater uptake of liposome encapsulated drug than of the free compound. Monocytes do not degrade the 3H-nor MDP that they have internalized and radiolabel is only slowly released from the cells. These observations suggest a pharmacokinetic basis for the use of lipid vesicles as a system for the **delivery** of immunomodulating drugs to monocytes.

=> D HIS L34-

(FILE 'BIOSIS' ENTERED AT 08:37:37 ON 25 SEP 96)
L34 11 S L12
L35 0 S L14
L36 10 S L17
L37 7 S L18
L38 23 S L34-L37

FILE 'HCAPLUS' ENTERED AT 08:42:45 ON 25 SEP 96
L39 27 S L12 OR L14 OR L17 OR L18

FILE 'BIOSIS' ENTERED AT 08:42:53 ON 25 SEP 96

=> D L34 1-11 BIB ABS;D L36 1-10 BIB ABS;D L37 1-7 BIB ABS

L34 ANSWER 1 OF 11 BIOSIS COPYRIGHT 1996 BIOSIS
AN 96:370395 BIOSIS
DN 99092751
TI Interaction of polyions with cell-mimetic species: Physico-chemical and biomedical aspects.
AU Kabanov V A; Yaroslavov A A; Sukhishvili S A
CS Dep. Polymer Sci., Fac. Chem., Moscow State Univ., Moscow 119899, Russia
SO Journal of Controlled Release 39 (2-3). 1996. 173-189. ISSN: 0168-3659
LA English
AB The possibility of recognition and discrimination of relatively large charged supermolecular objects (latex species) by an oppositely charged polyion is demonstrated using a suspension of carboxylated and protein-modified latex particles interacting with the high molecular mass linear **polycations** including those conjugated with the specific protein (alpha-chymotrypsin). The **polycations** are strongly adsorbed on the latex surface. Nevertheless, they are able to migrate between the latex species via occasional interparticle contacts. Finally, the interchanging **polycations** carrying the specific protein are fixed on those latex particles which carry the complementary protein receptor (trypsin inhibitor from soybean). The presence of other proteins does not hinder such interaction. The resulting effect is considered to mimic a physico-chemical aspect of recognition of **target cells** by macromolecules combined with relatively small molecular vector. Interaction of the **target cell** membrane with a **polycation** was simulated using **negatively** charged liposomes. It was found that **polycations** adsorbed on the surface of liquid liposomes can cause a significant charge asymmetry in the lipid bilayer due to transmembrane migration of **negatively** charged lipids from the inner to outer leaflet. At the same time the liposomal membrane integrity can be retained and adsorbed **polycations** can be replaced from the membrane by recomplexation with **polyanion** species. The established phenomena may be important for understanding the biological effects of **polycations**. **Negatively** charged liquid liposomes were also used to mimic interaction of cells with **DNA-polycation** and **DNA-cationic** surfactant complexes used to enhance plasmid **DNA** translocation. It was found that the complex of

DNA with the **polycation** carrying hydrophobic side groups interacted with the liposomes without dissociation and adsorbed on the liposome surface as a whole.

L34 ANSWER 2 OF 11 BIOSIS COPYRIGHT 1996 BIOSIS
AN 96:230304 BIOSIS
DN 98794433
TI Folate-targeted, **anionic** liposome-entrapped **polylysine**-condensed **DNA** for tumor cell-specific gene transfer.
AU Lee R J; Huang L
CS Lab. Drug Targeting, Dep. Pharmacol., Univ. Pittsburgh Sch. Med., Pittsburgh, PA 15261, USA
SO Journal of Biological Chemistry 271 (14). 1996. 8481-8487. ISSN: 0021-9258
LA English
AB We have developed a lipidic gene transfer vector, LPD-II, where **DNA** was first complexed to **polylysine** at a ratio of 1:0.75 (w/w) and then entrapped into folate-targeted pH-sensitive **anionic** liposomes composed of dioleoyl phosphatidylethanolamine (DOPE)/cholesteryl hemisuccinate/folate-polyethlene glycol-DOPE (6:4:0.01 mol/mol) via charge interaction. LPDII transfection of KB cells, a cell line overexpressing the tumor marker folate receptor, was affected by both the lipid to **DNA** ratio and the lipid composition. At low lipid to **DNA** ratios (eg. 4 and 6), LPDII particles were **positively** charged; transfection and cellular uptake levels were independent of the folate receptor and did not require a pH-sensitive lipid composition. Meanwhile, transfection and uptake of **negatively** charged LPDII particles, i.e. those with high lipid to **DNA** ratios (eg. 10 and 12), were folate receptor-dependent and required a pH-sensitive lipid composition. The transfection activity of LPDII was lost when the inverted cone-shaped DOPE was replaced by dioleoyl phosphatidylcholine. LPDII particles with lipid to **DNA** ratios of 4, 6, 10, and 12 were apprx 20-30 times more active than **DNA** cntdot 3-beta-(N-(N',N'-dimethylethane)carbamoyl)choleste rol **cationic** liposome complexes in KB cells and were much less cytotoxic. On the sucrose gradient, LPDII particles had a migration rate in between those of the free **DNA** and the **DNA** cntdot **polylysine** complex. An electron micrograph of LPDII showed a structure of spherical particles with a **positively** stained core enclosed in a lipidic envelope with a mean diameter of 74 +- 14 nm. This novel gene transfer vector may potentially be useful in gene therapy for tumor-specific **delivery**.

L34 ANSWER 3 OF 11 BIOSIS COPYRIGHT 1996 BIOSIS
AN 96:123176 BIOSIS
DN 98695311
TI Potentiation of **cationic** liposome-mediated gene **delivery** by **polycations**.
AU Gao X; Huang L
CS Dep. Pharmacol., Univ. Pittsburgh Sch. Medicine, Pittsburgh, PA 15261, USA
SO Biochemistry 35 (3). 1996. 1027-1036. ISSN: 0006-2960
LA English
AB We discovered that several high molecular weight **cationic** polymers, such as **poly**(L-**lysine**) and protamine,

can enhance the transfection efficiency of several types of **cationic** liposomes by 2-28-fold in a number of cell lines in vitro. Small **polycations** such as spermine and a **cationic** decapeptide derived from SV40 T-antigen were only moderately active. The addition of **poly**(L-**lysine**) and protamine dramatically reduced the particle size of the complex formed between **DNA** and **cationic** liposomes and rendered **DNA** resistant to the nuclease activity. The complexes composed of **DNA**, **poly**(L-**lysine**), and **cationic** lipids were purified from an excess of free liposomes with sucrose gradient ultracentrifugation. Purified complex formed at low **cationic** liposome ratio was poor in lipid content and only had weak transfection activity. Addition of free liposome to the purified complex significantly enhanced the transfection activity. In contrast, complexes formed at a higher initial ratio of liposome to **DNA** had a higher lipid content and were highly active in transfection; the activity was about 3-9-fold more active than the corresponding complex before purification. **Negative** stain EM studies revealed that the most active complexes prepared from 40 nmol of lipid, 0.5 mu-g of **poly**(L-**lysine**), and 1 mu-g of **DNA** and purified by gradient ultracentrifugation were spherical, electron dense, small (lt 100 nm in diameter) particles, and some of them were associated with lipid membranes. These highly active, stable, small-sized lipid/**poly**(L-**lysine**)/**DNA** complexes represent a new class of nonviral gene **delivery** vehicles that might be useful in gene therapy.

L34 ANSWER 4 OF 11 BIOSIS COPYRIGHT 1996 BIOSIS
AN 95:437107 BIOSIS
DN 98451407
TI A versatile vector for gene and **oligonucleotide** transfer into cells in culture and in vivo: Polyethylenimine.
AU Boussif O; Lezoualc'h F; Zanta M A; Mergny M D; Scherman D; Demenix B; Behr J-P
CS Lab. Chim. Genet., Unite Rech. Assoc. 1386 Cent. Natl. Rech. Sci., Fac. Pharm., F-67401 Illkirch, France
SO Proceedings of the National Academy of Sciences of the United States of America 92 (16). 1995. 7297-7301. ISSN: 0027-8424
LA English
AB Several **polycations** possessing substantial buffering capacity below physiological pH, such as lipopolyamines and polyamidoamine **polymers**, are efficient transfection agents per se sbd i.e., without the addition of **cell targeting** or membrane-disruption agents. This observation led us to test the **cationic polymer** polyethylenimine (PEI) for its gene **delivery** potential. Indeed, every third atom of PEI is a protonable amino nitrogen atom, which makes the **polymeric** network an effective "proton sponge" at virtually any pH. Luciferase reporter gene transfer with this **polycation** into a variety of cell lines and primary cells gave results comparable to, or even better than, lipopolyamines. Cytotoxicity was low and seen only at concentrations well above those required for optimal transfection. **Delivery** of **oligonucleotides** into embryonic neurons was followed by using a fluorescent probe. Virtually all neurons showed nuclear labeling, with no toxic effects. The optimal PEI **cation/anion** balance for in vitro transfection is only slightly on the

cationic side, which is advantageous for in vivo **delivery**. Indeed, intracerebral luciferase gene transfer into newborn mice gave results comparable (for a given amount of **DNA**) to the in vitro transfection of primary rat brain endothelial cells or chicken embryonic neurons. Together, these properties make PEI a promising vector for gene therapy and an outstanding core for the design of more sophisticated devices. Our hypothesis is that its efficiency relies on extensive lysosome buffering that protects **DNA** from nuclease degradation, and consequent lysosomal swelling and rupture that provide an escape mechanism for the PEI/**DNA** particles.

L34 ANSWER 5 OF 11 BIOSIS COPYRIGHT 1996 BIOSIS
AN 95:392935 BIOSIS
DN 98407235
TI Natural Killer (NK) Activity in Human Responders and Nonresponders to Stimulation by Anti-CD16 Antibodies.
AU Galatiuc C; Gherman M; Metes D; Sulica A; Deleo A; Whiteside T L; Herberman R B
CS Cent. Immunol., Bucharest, Romania
SO Cellular Immunology 163 (2). 1995. 167-177. ISSN: 0008-8749
LA English
AB Various anti-Fc-gamma-RIII (CD16) monoclonal antibodies (mAbs) are shown here to have **positive** or **negative** modulatory effects on human NK cells. Thus, 3G8 mAb (IgG-1) triggered a dose-dependent augmentation of NK activity in 67% (23/34) of individuals tested, who were designated as responders. All four IgG-1 anti-CD16 mAb tested (BL-LGL/1, B73.1, Leu11c, and 3G8) were stimulatory for NH cells isolated from responders, whereas six non-IgG-1 anti-CD16 mAbs were either inhibitory or had no significant effects on NK activity. The upregulation of NK activity in responders was not attributable to an increase in either the conjugate formation or the **delivery** of the lethal hit to **target cells**. This mAb-mediated up-regulation of NK activity was shown to be associated with a recycling capacity higher than that of controls and with enhanced release of cytokines by activated NK cells. Anti-CD16 mAb inhibited binding of either monomeric or **polymeric** IgG to Fc-gamma-RIIIA on NK cells. Also, mAb 3G8 or its F(ab')-2 fragments decreased or reversed inhibition of NK activity induced by monomeric IgG (mIgG). Our data indicate that regulation of NK activity via the Fc-gamma-RIIIA is influenced by dose-dependent interactions between cytophilic mIgG and anti-CD16 mAb of IgG-1 isotype.

L34 ANSWER 6 OF 11 BIOSIS COPYRIGHT 1996 BIOSIS
AN 94:484995 BIOSIS
DN 97497995
TI Rapid protection against human immunodeficiency virus type 1 (HIV-1) replication mediated by high efficiency non-retroviral **delivery** of genes interfering with HIV-1 tat and gag.
AU Lori F; Lisziewicz J; Smythe J; Cara A; Bunnag T A; Curiel D; Gallo R C
CS Lab. Tumor Cell Biol., Natl. Cancer Inst., NIH, Bethesda, MD 20892, USA
SO Gene Therapy 1 (1). 1994. 27-31.
LA English
AB Efficient transduction of inhibitory genes is a critical requirement in the development of a gene therapy strategy against human

immunodeficiency virus type 1 (HIV-1). Commonly used systems based on retrovirus-mediated gene **delivery** are characterized by low efficiency gene transfer into the **target cell**. Genes were transduced in the absence of cell selection into 60-90% of human CD4+ cells by using a novel technique that allows high efficiency gene transfer mediated by adenoviruses coupled with **DNA-polylysine** complexes. Protection of these cells against HIV-1 acute infection was evaluated by transducing them with three different inhibitory genes which interfere with HIV-1 replication at separate level (**polymeric** Tat activation response element (TAR) decoy, dominant-**negative** mutant of the gag gene and antisense sequences of the gag gene) and subsequent challenging with HIV-1. The **polymeric** TAR decoy inhibited HIV-1 replication over 95%. Both the dominant-**negative** mutant and the antisense sequence of the gag gene were less potent inhibitors than the **polymeric**-TAR decoy. Combinations of either **polymeric**-TAR with dominant-**negative** mutant or antisense of the gag gene synergistically enhanced the inhibitory effects of the single genes. These data suggest that the combination of a highly efficient transduction technique with effective HIV-1 inhibitory genes confers rapid protection against HIV-1 acute infection in vitro.

L34 ANSWER 7 OF 11 BIOSIS COPYRIGHT 1996 BIOSIS
AN 93:230038 BIOSIS
DN BA95:121213
TI TARGETED TRANSFECTION AND EXPRESSION OF HEPATITIS B VIRAL **DNA** IN HUMAN HEPATOMA CELLS.
AU LIANG T J; MAKDISI W J; SUN S; HASEGAWA K; ZHANG Y; WANDS J R; WU C H; WU G Y
CS GASTROINTESTINAL UNIT, MASS. GEN. HOSP., JACKSON 812, BOSTON, MA 02114, USA.
SO J CLIN INVEST 91 (3). 1993. 1241-1246. CODEN: JCINAO ISSN: 0021-9738
LA English
AB A soluble **DNA** carrier system consisting of an asialoglycoprotein covalently linked to **poly**-L-**lysine** was used to bind **DNA** and **deliver** hepatitios B virus (HBV) **DNA** constructs to asialoglycoprotein receptor-**positive** human hepatoma cells. 4 d after transfection with surface or core gene expression constructs, HBsAg and HBeAg in the media were measured to be 16 ng/ml and 32 U/ml per 107 cells, respectively. Antigen production was completely inhibited by the addition of an excess of asialoorosomucoid. On the other hand, asialoglycoprotein receptor-**negative** human hepatoma cells, SK-Hep1, did not produce any viral antigens under indentical conditions after incubation with HBV **DNA** complexed to a conjugate composed of asialoorosomucoid and **poly**-L-**lysine**. Using a complete HBV genome construct, HBsAg and HBeAg levels reached 16 ng/ml and 16 U/ml per 107 cells, respectively. Northern blots revealed characteristic HBV RNA transcripts including 3.5-, 2.4-, and 2.1-kb fragments. Intracellular and extracellular HBV **DNA** sequences including relaxed circular, linear and single stranded forms were detected by Southern blot hybridization. Finally, 42-nm Dane particles purified from the spent culture medium were visualized by electron microscopy. This study demonstrate that a targetable **DNA** carrier system can transfect HBV **DNA** in vitro resulting in the production of complete HBV virions.

L34 ANSWER 8 OF 11 BIOSIS COPYRIGHT 1996 BIOSIS
AN 92:523490 BIOSIS
DN BA94:131565
TI CHIMERIC **DNA**-RNA HAMMERHEAD RIBOZYMES HAVE ENHANCED IN-VITRO CATALYTIC EFFICIENCY AND INCREASED STABILITY IN-VIVO.
AU TAYLOR N R; KAPLAN B E; SWIDERSKI P; LI H; ROSSI J J
CS DEP. MOLECULAR GENETICS, BECKMAN RESEARCH INSTITUTE CITY OF HOPE, DUARTE, CALIF. 91010.
SO NUCLEIC ACIDS RES 20 (17). 1992. 4559-4565. CODEN: NARHAD ISSN: 0305-1048
LA English
AB Subsequent to the discovery that RNA can have site specific cleavage activity, there has been a great deal of interest in this design and testing of trans-acting catalytic RNAs as both surrogate genetic tools and as therapeutic agents. We have been developing catalytic RNAs or ribozymes with target specificity for HIV-1 RNA and have been exploring chemical synthesis as one method for their production. To this end, we have chemically synthesized and experimentally analyzed chimeric catalysts consisting of **DNA** in the nonenzymatic portions, and RNA in the enzymatic core of hammerhead type ribozymes. Substitutions of **DNA** for RNA in the various stems of a hammerhead ribozyme have been analyzed in vitro for kinetic efficiency. One of the chimeric ribozymes used in this study, which harbors 24 bases of **DNA** capable of base-pairing interactions with an HIV-1 gag target, but maintains RNA in the catalytic center and in stem-loop II, has a sixfold greater kcat value than the all RNA counterpart. This increased activity appears to be the direct result of enhanced product dissociation. Interestingly, a chimeric ribozyme in which stem-loop II (which divides the catalytic core) is comprised of **DNA**, exhibited a marked reduction in cleavage activity, suggesting that **DNA** in this region of the ribozyme can impart a **negative** effect on the catalytic function of the ribozyme. **DNA**-RNA chimeric ribozymes transfected by **cationic** liposomes into human T-lymphocytes are more stable than their all-RNA counterparts. Enhanced catalytic turnover and stability in the absence of a significant effect on Km make chimeric ribozymes favorable candidates for therapeutic agents.

L34 ANSWER 9 OF 11 BIOSIS COPYRIGHT 1996 BIOSIS
AN 92:502070 BIOSIS
DN BA94:120595
TI CLASS I RESTRICTED CTL RECOGNITION OF A SOLUBLE PROTEIN **DELIVERED** BY LIPOSOMES CONTAINING LIPOPHILIC POLYLYSINES.
AU NAIR S; ZHOU X; HUANG L; ROUSE B T
CS DEP. MICROBIOL., UNIV. TENN., KNOXVILLE, TENN. 37996-0845, USA.
SO J IMMUNOL METHODS 152 (2). 1992. 237-243. CODEN: JIMMBG ISSN: 0022-1759
LA English
AB CD8+ cytotoxic lymphocytes recognize peptides derived from endogenous antigens complexed with class I major histocompatibility complex while CD4+ helper cells recognize peptides from exogenous antigens bound to class II MHC molecules. A soluble protein can be introduced into the class I pathway of antigen processing and presentation using an appropriate vehicle to **deliver** the antigen into the cytosol. **Cationic** liposomes containing lipophilic **polylysine** readily form complexes with an **anionic**,

soluble protein ovalbumin. Mouse thymoma EL4 cells incubated with such complexes can be sensitized for killing by OVA-specific CTL effector **cells**. This method of **target** sensitization by a soluble antigen is more sensitive than the osmotic loading method previously reported.

L34 ANSWER 10 OF 11 BIOSIS COPYRIGHT 1996 BIOSIS
AN 90:176425 BIOSIS
DN BA89:93595
TI TRANSFER OF PREFORMED TERMINAL C5B-9 COMPLEMENT COMPLEXES INTO THE OUTER MEMBRANE OF VIABLE GRAM-**NEGATIVE** BACTERIA EFFECT ON VIABILITY AND INTEGRITY.
AU TOMLINSON S; TAYLOR P W; LUZIO J P
CS ADDENBROOKE'S HOSP., HILLS RD., CAMBRIDGE CB2 2QR, UK.
SO BIOCHEMISTRY 29 (7). 1990. 1852-1860. CODEN: BICHAW ISSN: 0006-2960
LA English
AB An efficient fusion system between Gram-**negative** bacteria and liposomes incorporating detergent-extracted C5b-9 complexes has been developed that allows **delivery** of preformed terminal complexes to the cell envelope (Tomlinson et al., 1989b). Fusion of Salmonella minnesota Re595 and Escherichia coli 17 with C5b-9-incorporated liposomes resulted in the transfer of 1900 C5b-9 complexes to each **target** bacterial **cell**. No loss in viability of bacteria was observed following fusion, even though the deposition of 900 complexes onto the envelope following exposure to lysozyme-free serum effected a greater than 99% loss of viability. Increased sensitivity to antibiotics normally excluded from the cell by an integral outer membrane (OM), as well as the ability of the chromogenic substrate PADAC to gain access to periplasmically located .beta.-lactamase, indicated that transferred C5b-9 complexes functioned as water-filled channels through the OM. A similar conclusion was drawn from measurements demonstrating the uptake by cells of the lipophilic **cation** tetraphenylphosphonium (bromide), a result further indicating that the membrane potential across the cytoplasmic membrane was maintained following C5b-9 transfer to the OM. Examination of S. minnesota Re595 by electron microscopy revealed no obvious difference between cells exposed to lethal concentrations of lysozyme-free serum and cells following fusion with C5b-9-incorporated liposomes. These data suggest either that there are critical sites in the OM to which liposome-**delivered** C5b-9 complexes are unable to gain access or that bacterial cell death is related to events occurring during **polymerization** of C9 on the cell surface.

L34 ANSWER 11 OF 11 BIOSIS COPYRIGHT 1996 BIOSIS
AN 90:6658 BIOSIS
DN BA89:6658
TI THE INTRACELLULAR RELEASE OF METHOTREXATE FROM A SYNTHETIC DRUG CARRIER SYSTEM TARGETED TO FC RECEPTOR-BEARING CELLS.
AU SHEN W-C; DU X; FEENER E P; RYSER H J-P
CS UNIV. SOUTHERN CALIF. SCH. PHARM., LOS ANGELES, CALIF. 90033.
SO J CONTROLLED RELEASE 10 (1). 1989. 89-96. CODEN: JCREEC ISSN: 0168-3659
LA English
AB Methotrexate was conjugated to trinitrophenyl (TNP)-labeled **poly**(D-**lysine**) either directly, or through a disulfide or a triglycine spacer. Conjugates were complexed with heparin and anti-TNP antiserum and tested for their growth inhibitory

effects in cultured cells. When tested in Fc receptor-**positive** WEHI-3 cells, the anti-TNP immune complex of the conjugate with the disulfide spacer was more effective than that with the triglycine spacer. It had no effect on the growth of Fc receptor-**negative** L-929 cells. Without spacer, the direct conjugate was ineffective in both cell lines. The growth inhibitory effect of the drug-containing immune complexes was partially abolished in presence of an irrelevant, drug-free immune complex. NH4Cl at 3 mM did not increase the cytotoxicity of the disulfide conjugate complexes, and decreased the effect of the triglycine conjugate complexes. These findings suggest that: (1) hapten-**polymer** conjugates can be used as drug carriers for **targeting** Fc receptor-bearing **cells** when given with an anti-hapten antibody, (2) disulfide spacers are effectively cleaved following Fc receptor-mediated endocytosis of a drug-carrying immune complex, (3) disulfide spacers are most likely to be cleaved in compartments other than endosomes or lysosomes.

L36 ANSWER 1 OF 10 BIOSIS COPYRIGHT 1996 BIOSIS
AN 96:370395 BIOSIS
DN 99092751
TI Interaction of polyions with cell-mimetic species: Physico-chemical and biomedical aspects.
AU Kabanov V A; Yaroslavov A A; Sukhishvili S A
CS Dep. Polymer Sci., Fac. Chem., Moscow State Univ., Moscow 119899, Russia
SO Journal of Controlled Release 39 (2-3). 1996. 173-189. ISSN: 0168-3659
LA English
AB The possibility of recognition and discrimination of relatively large charged supermolecular objects (latex species) by an oppositely charged polyion is demonstrated using a suspension of carboxylated and protein-modified latex particles interacting with the high molecular mass linear **polycations** including those conjugated with the specific protein (alpha-chymotrypsin). The **polycations** are strongly adsorbed on the latex surface. Nevertheless, they are able to migrate between the latex species via occasional interparticle contacts. Finally, the interchanging **polycations** carrying the specific protein are fixed on those latex particles which carry the complementary protein **receptor** (trypsin inhibitor from soybean). The presence of other proteins does not hinder such interaction. The resulting effect is considered to mimic a physico-chemical aspect of recognition of **target cells** by macromolecules combined with relatively small molecular vector. Interaction of the **target cell** membrane with a **polycation** was simulated using **negatively** charged liposomes. It was found that **polycations** adsorbed on the surface of liquid liposomes can cause a significant charge asymmetry in the lipid bilayer due to transmembrane migration of **negatively** charged lipids from the inner to outer leaflet. At the same time the liposomal membrane integrity can be retained and adsorbed **polycations** can be replaced from the membrane by recomplexation with **polyanion** species. The established phenomena may be important for understanding the biological effects of **polycations**. **Negatively** charged liquid liposomes were also used to mimic interaction of cells

with DNA-**polycation** and DNA-**cationic** surfactant complexes used to enhance plasmid DNA translocation. It was found that the complex of DNA with the **polycation** carrying hydrophobic side groups interacted with the liposomes without dissociation and adsorbed on the liposome surface as a whole.

L36 ANSWER 2 OF 10 BIOSIS COPYRIGHT 1996 BIOSIS
AN 95:392935 BIOSIS
DN 98407235
TI Natural Killer (NK) Activity in Human Responders and Nonresponders to Stimulation by Anti-CD16 Antibodies.
AU Galatiuc C; Gherman M; Metes D; Sulica A; Deleo A; Whiteside T L; Herberman R B
CS Cent. Immunol., Bucharest, Romania
SO Cellular Immunology 163 (2). 1995. 167-177. ISSN: 0008-8749
LA English
AB Various anti-Fc-gamma-RIII (CD16) monoclonal antibodies (mAbs) are shown here to have **positive** or **negative** modulatory effects on human NK cells. Thus, 3G8 mAb (IgG-1) triggered a dose-dependent augmentation of NK activity in 67% (23/34) of individuals tested, who were designated as responders. All four IgG-1 anti-CD16 mAb tested (BL-LGL/1, B73.1, Leu11c, and 3G8) were stimulatory for NH cells isolated from responders, whereas six non-IgG-1 anti-CD16 mAbs were either inhibitory or had no significant effects on NK activity. The upregulation of NK activity in responders was not attributable to an increase in either the conjugate formation or the **delivery** of the lethal hit to **target cells**. This mAb-mediated up-regulation of NK activity was shown to be associated with a recycling capacity higher than that of controls and with enhanced release of cytokines by activated NK cells. Anti-CD16 mAb inhibited binding of either monomeric or polymeric IgG to Fc-gamma-RIIIA on NK cells. Also, mAb 3G8 or its F(ab')-2 fragments decreased or reversed inhibition of NK activity induced by monomeric IgG (mIgG). Our data indicate that regulation of NK activity via the Fc-gamma-RIIIA is influenced by dose-dependent interactions between cytophilic mIgG and anti-CD16 mAb of IgG-1 isotype.

L36 ANSWER 3 OF 10 BIOSIS COPYRIGHT 1996 BIOSIS
AN 94:486267 BIOSIS
DN 97499267
TI Epidermal growth factor **receptors** in human breast carcinoma **cells**: A potential selective **target** for transforming growth factor alpha-Pseudomonas exotoxin 40 fusion protein.
AU Arteaga C L; Hurd S D; Dugger T C; Winnier A R; Robertson J B
CS Div. Med. Oncol., Vanderbilt Univ., 22nd Ave. South, 1956 TVC, Nashville, TN 37232-5536, USA
SO Cancer Research 54 (17). 1994. 4703-4709. ISSN: 0008-5472
LA English
AB Epidermal growth factor (EGF) **receptors** are expressed in high levels by some poor prognosis breast tumors. We have examined the cytotoxic effect of the tumor growth factor alpha (TGF-alpha)-DELTA-Cys-Pseudomonas exotoxin (PE40) recombinant fusion protein on normal and tumorigenic human breast epithelial cells in vitro and in vivo. The MDA-468, MDA-231, BT-20, and MCF-7-ADR estrogen **receptor-negative**, EGF **receptor** -rich breast cancer lines were exquisitely sensitive in vitro to

TGF-alpha-DELTA-Cys-PE40 with a 50% inhibitory concentration of ltoreq 0.02 nM. The estrogen **receptor-positive**, low EGF **receptor** MCF-7, ZR75-1, and T47D cells were less sensitive to the fusion toxin with a 50% inhibitory concentration of gt 0.2 nM. The nontumorigenic cell lines 184, 184A1, and 184B5 were relatively resistant to TGF-alpha-DELTA-Cys-PE40 despite exhibiting high levels of EGF **receptors**. Continuous i.p. administration of TGF-alpha-DELTA-Cys-PE40 via an osmotic minipump at a dose of 0.4 mu-g/g/day over 7 days inhibited MDA-468, MA-231, and BT-20 but not MCF-7 tumor growth in female athymic mice. Host tissue toxicity was not observed with this dose of TGF-alpha-DELTA-Cys-PE40. Mixed MDA-468/MCF-7 tumors were established in nude mice after coinoculation of both cell types in estrogen-supplemented animals. EGF **receptor** immunohistochemistry and immunoblot procedures indicated that TGF-alpha-PE40 eliminated the MDA-468 cells while sparing the adjacent MCF-7 cells. By immunoblot, EGF **receptors** were consistently more abundant in tumor tissue than in adjacent nontumor tissue from the same mastectomy specimen (n = 7). These data support the notion that EGF **receptors** can be selectively targeted in human breast cancer cells for the **delivery** of antitumor agents. Further clinical studies with TGF-alpha-DELTA-Cys-PE40 and other chimeric toxins using the same **cellular target** will address this possibility.

L36 ANSWER 4 OF 10 BIOSIS COPYRIGHT 1996 BIOSIS
AN 94:438511 BIOSIS
DN 97451511
TI Signal requirement for induction of MHC-unrestricted antitumor cytotoxicity of human T cell CD4+-CD8+ subpopulations.
AU Zhu H-G; Klein-Franke A; Anderer F A
CS Friedrich-Miescher-Lab. Max-Planck-Gesellschaft, Spemannstrasse 37/39, D 72076 Tuebingen, GER
SO Anticancer Research 14 (3A). 1994. 953-961. ISSN: 0250-7005
LA English
AB The role of cosignalling in the generation of MHC-unrestricted cytotoxicity of T cells was studied with CD4+ and CD8+ sub-populations highly purified (gt 98%) by immunomagnetic cell sorting using OKT4 mab, Dynal anti-CD4 mab, OKT8 mab, Dynal anti-CD8 mab, and OKT3 mab. Cytotoxicity was determined in 4 h cytotoxicity assays against K562 tumor cells known to lack expression of MHC class 1 and class 2 antigens, thus avoiding interference with anti-CD4- or anti-CD8-mediated signalling. Signal transfer was induced via CD4, CD8, CD3, IL-2 **receptor** and RG **receptor** specifically interacting with a plant rhamnogalacturonan (RG). In CD8+ cells, the first signal **delivered** by the sorting mab (immobilized OKT8 or Dynal anti-CD8 or OKT3) only induced low MHC-unrestricted cytotoxicity but committed the cells to develop largely enhanced cytolytic potential upon stimulation with a second (IL-2 or RG) or third (OKT3, IL-2, RG) signal. The highest cytolytic potential was achieved by cumulative signalling via CD8, CD3, IL-2 **receptor** and RG **receptor**. The generation of MHC-unrestricted cytotoxicity of CD8+ cells correlated with increased effector **cell/target cell** conjugate formation. In CD4+ cells, OKT4 as sorting mab induced very low cytolytic potential, and a moderate committment to IL-2 signals but a stronger one to RG signals, yielding further cytotoxicity enhancement. The highest cytolytic potential was obtained by cumulative signalling via CD4, IL-2 **receptor** and RG

receptor. Dynal anti-CD4 mab was inefficient and OKT3, as sorting mab of CD4 + cells from CD8-depleted PNAC, appeared to block subsequent OKT4-induced generation of MHC-unrestricted cytotoxicity by **delivering** a **negative** signaL Immobilized OKT3 as second signal present in cultures of OKT4-sorted CD4+ cells was inefficient. Surprisingly, soluble OKT3 together with IL2 **delivered** a **positive** signal in cultures of OKT4-sorted CD4+ cells.

L36 ANSWER 5 OF 10 BIOSIS COPYRIGHT 1996 BIOSIS
AN 93:456494 BIOSIS
DN BA96:101394
TI HUMAN T-CELL LEUKEMIA VIRUS TYPE I-INDUCED PROLIFERATION OF HUMAN IMMATURE CD2-**POSITIVE** CD3-**NEGATIVE** THYMOCYTES.
AU MAGUER V; CASSE-RIPOLL H; GAZZOLO L; DODON M D
CS IMMUNO-VIROL. MOL. ET CELL., UMBR30, CENTRE NATIONAL DE LA RECHERCHE SCIENTIFIQUE, UNIV. CLAUDE BERNARD LYON I, FAC. DE MED. A. CARRELL, 69372 LYON CEDEX 08, FR.
SO J VIROL 67 (9). 1993. 5529-5537. CODEN: JOVIAM ISSN: 0022-538X
LA English
AB The mitogenic activity of human T-cell leukemia virus type I (HTLV-I) is triggering the proliferation of human resting T lymphocytes through the induction of the interleukin-2 (IL-2)/IL-2 **receptor** autocrine loop. This HTLV-I-induced proliferation was found to be mainly mediated by the CD2 T-cell antigen, which is first expressed on double-**negative** lymphoid precursors after colonization of the thymus. Thus, immature thymocytes express the CD2 antigen before that of the CD3-TCR complex. We therefore investigated the responsiveness of these CD2+CD3- immature thymocytes and compared it with that of unseparated thymocytes, containing a majority of the CD2+CD3+ mature thymocytes, and that of the CD2-CD3prothymocytes. Both immature and unseparated thymocytes were incorporating [3H]thymidine in response to the virus, provided that they were cultivated in the presence of submitogenic doses of phytohemagglutinin. In contrast, the prothymocytes did not proliferate. Downmodulation of the CD2 molecule by incubating unseparated and immature thymocytes with a single anti-CD2 monoclonal antibody inhibited the proliferative response to HTLV-I. These results clearly underline that the expression of the CD2 molecule is exclusively required in mediating the proliferative response to the synergistic effect of phytohemagglutinin and HTLV-I. Immature thymocytes treated with a pair of anti-CD2 monoclonal antibodies were shown to proliferate in response to HTLV-I, even in the absence of exogenous IL-2. We further verified that the proliferation of human thymocytes is consecutive to the expression of IL-2 **receptors** and the synthesis of IL-2. These observations provide evidence that the mitogenic stimulus **delivered** by HTLV-I is more efficient than that provided by other conventional mitogenic stimuli, which are unable to trigger the synthesis of endogenous IL-2. Collectively, these results show that the mitogenic activity of HTLV-I is able to trigger the proliferation of cells which are at an early stage of T-cell development. They might therefore represent **target cells** in which HTLV-I infection could favor the initiation of the multistep lymphoproliferative process leading to adult T-cell leukemia.

L36 ANSWER 6 OF 10 BIOSIS COPYRIGHT 1996 BIOSIS
AN 91:478931 BIOSIS

DN BA92:112691
TI T-CELL-**RECEPTOR**-INDEPENDENT ACTIVATION OF CYTOLYTIC ACTIVITY OF CYTOTOXIC T LYMPHOCYTES MEDIATED THROUGH CD44 AND GP90M-E-L-14.
AU SETH A; GOTE L; NAGARKATTI M; NAGARKATTI P S
CS DEP. BIOL., DIV. MICROBIOL. IMMUNOL., VIRGINIA POLYTECHNIC INST. STATE UNIVERSITY, BLACKSBURG, VA. 24061.
SO PROC NATL ACAD SCI U S A 88 (17). 1991. 7877-7881. CODEN: PNASA6 ISSN: 0027-8424
LA English
AB CD44 is a transmembrane glycoprotein found on a variety of cells including those of myeloid and lymphoid origin. CD44 is highly conserved among various species and is involved in the homing of lymphocytes and monocytes to lymph nodes, Peyer's patches, and sites of inflammation. In the present study, we demonstrate that monoclonal antibody (mAb) 9F3, directed against phagocytic glycoprotein 1 (CD44) expressed on cytotoxic T lymphocytes (CTLs), can trigger the lytic activity of CTLs and redirect CTL-mediated lysis to antigen-**negative** Fc **receptor-positive** **target cells**. Similar redirected lysis was also inducible using mAb MEL-14, directed against the lymphocyte homing **receptor** for endothelium (gp90MEL-14). The redirected lysis induced by mAbs 9F3 and MEL-14 was similar to that induced by mAbs against the .alpha..beta. T-cell **receptor** or CD3. In contrast, mAbs directed against CD8, CD45R, and CD11a (LFA-1, lymphocyte function-associated antigen 1) failed to evoke lytic activity. The current study demonstrates that CD44 and gp90MEL-14 molecules in additon to participating in T-cell homing and adhesion, may play a major role in **delivering** the transmembrane signal to the CTL that triggers the lytic activity, even when the T-cell **receptor** is not occupied. Such a mechanism may account for the nonspecific tissue damage seen at sites of CTL-mediated inflammation.

L36 ANSWER 7 OF 10 BIOSIS COPYRIGHT 1996 BIOSIS
AN 90:6658 BIOSIS
DN BA89:6658
TI THE INTRACELLULAR RELEASE OF METHOTREXATE FROM A SYNTHETIC DRUG CARRIER SYSTEM TARGETED TO FC **RECEPTOR**-BEARING CELLS.
AU SHEN W-C; DU X; FEENER E P; RYSER H J-P
CS UNIV. SOUTHERN CALIF. SCH. PHARM., LOS ANGELES, CALIF. 90033.
SO J CONTROLLED RELEASE 10 (1). 1989. 89-96. CODEN: JCREEC ISSN: 0168-3659
LA English
AB Methotrexate was conjugated to trinitrophenyl (TNP)-labeled poly(D-lysine) either directly, or through a disulfide or a triglycine spacer. Conjugates were complexed with heparin and anti-TNP antiserum and tested for their growth inhibitory effects in cultured cells. When tested in Fc **receptor-positive** WEHI-3 cells, the anti-TNP immune complex of the conjugate with the disulfide spacer was more effective than that with the triglycine spacer. It had no effect on the growth of Fc **receptor-negative** L-929 cells. Without spacer, the direct conjugate was ineffective in both cell lines. The growth inhibitory effect of the drug-containing immune complexes was partially abolished in presence of an irrelevant, drug-free immune complex. NH4Cl at 3 mM did not increase the cytotoxicity of the disulfide conjugate complexes, and decreased the effect of the triglycine conjugate

complexes. These findings suggest that: (1) hapten-polymer conjugates can be used as drug carriers for **targeting** Fc **receptor**-bearing **cells** when given with an anti-hapten antibody, (2) disulfide spacers are effectively cleaved following Fc **receptor**-mediated endocytosis of a drug-carrying immune complex, (3) disulfide spacers are most likely to be cleaved in compartments other than endosomes or lysosomes.

L36 ANSWER 8 OF 10 BIOSIS COPYRIGHT 1996 BIOSIS
AN 89:127192 BIOSIS
DN BA87:61845
TI IMMUNOSPECIFIC TARGETING OF CYTOSINE ARABINONUCLEOSIDE-CONTAINING LIPOSOMES TO THE IDIOTYPE ON THE SURFACE OF A MURINE B-CELL TUMOR IN-VITRO AND IN-VIVO.
AU BANKERT R B; YOKOTA S; GHOSH S K; MAYHEW E; JOU Y-H
CS DEP. MOL. IMMUNOL., ROSWELL PARK MEML. INST., 666 ELM ST., BUFFALO, N.Y. 14263.
SO CANCER RES 49 (2). 1989. 300-308. CODEN: CNREA8 ISSN: 0008-5472
LA English
AB A new tumor model is described that is suitable for the evaluation of antibody-directed drug-**delivery** protocols and a modification in the procedure for covalently coupling antibody to the surface of drug-containing liposomes is presented. These immunospecific liposomes containing cytosine arabinonucleoside (Ara-C) have been tested in vitro and in vivo for their ability to kill a B-**cell** tumor. The **target** of the immunospecific Ara-C liposomes is the idiotype associated with an antigen-specific immunoglobulin **receptor** on the cell surface or a murine B-cell hybrid (2C3). Affinity-purified antibodies specific for the idiotype were covalently coupled to modified lipid on the surface of the large unilamelar liposomes containing drug. These liposomes were shown to kill idiotype-**positive** 2C3 cells in vitro, but not idiotype-**negative** variants of this same cell line. It was also established in vitro that the drug-containing liposomes were at least 40 times more efficient than free Ara-C in the killing of the tumor cells. The 2C3 tumor was also propagated in vivo following the i.p. administration of tumor cells. The tumor grew initially as multiple foci within the peritoneum and subsequently spread to the spleen. Tumor-bearing mice were treated either with free Ara-C or with immunospecific liposomes containing Ara-C. Tumor growth in the primary tumor nodules and in the spleen was monitored by the administration of bromodeoxyuridine to the tumor-bearing animals followed by the immunofluroescent staining of cells will a monoclonal anti-bromodeoxyuridine antibody to estimate the proportion of cells in S phase. Our data from five out of seven animals experiments shows that the immunospecific-Ara-C liposomes, but not free drug, reduced tumor growth in the spleen. However, neither the liposomes containing drug nor the free drug were able to alter the growth of the primary tumor nodules growing in the peritoneal cavity. These results suggest that immunospecific-Ara-C containing liposomes may be useful in conjunction with other cytoreductive protocols in controlling tumor growth or preventing the spread of the tumor to other sites, but that immunospecific-Ara-C containing liposomes by themselves are not likely to eliminate an established tumor in vivo. We also demonstrate here that the administration of immunospecific-Ara-C containing liposomes in an animal having high levels of circulating tumor-associated antigen (i.e., IgG containing the idiotype) represents a potential clinically

relevant hazard which must be considered when designing antibody-directed drug-**delivery** protocols.

L36 ANSWER 9 OF 10 BIOSIS COPYRIGHT 1996 BIOSIS
AN 88:418308 BIOSIS
DN BA86:80920
TI ANTI-L3T4 ANTIBODY INHIBITS THE LYSIS OF H-2 CLASS II ANTIGEN-**NEGATIVE TARGET CELLS** BY L3T4-**POSITIVE** CYTOTOXIC T LYMPHOCYTES.
AU MACPHAIL S; STUTMAN O
CS MEMORIAL SLOAN-KETTERING CANCER CENT., 1275 YORK AVE., NEW YORK, N.Y. 10021.
SO PROC NATL ACAD SCI U S A 85 (14). 1988. 5205-5209. CODEN: PNASA6 ISSN: 0027-8424
LA English
AB Anti-L3T4 monoclonal antibodies inhibit the cytotoxic activity of L3T4+ cytotoxic T lymphocytes specific for H-2 class I antigens. The P815 **target cells** used to detect this population of murine cytolytic cells are shown by immunofluorescence, radioimmunoprecipitation, and RNA blot analysis not to express H-2 class II protein or mRNA. Contrary to previously proposed models regarding its function, we conclude that the L3T4 molecule is involved at some stage of the lytic interaction between the class I-specific L3T4+ effector **cell** and its **target cell** by a mechanism for which there is not an obligatory requirement for H-2 class II antigen expression by the **target cell**. L3T4 may be an early component of the system that transduces the activation signal from the T-cell **receptor** complex to the cytoplasm, a cell-surface **receptor** for a yet undefined natural ligand that **delivers** a **negative** signal to the killer T cell, or it may modulate the avidity of the antigen-specific T-cell **receptor** through a direct physical association with it.

L36 ANSWER 10 OF 10 BIOSIS COPYRIGHT 1996 BIOSIS
AN 80:280304 BIOSIS
DN BA70:72800
TI **RECEPTOR** MEDIATED ENDOCYTOSIS OF ANTIBODY OPSONIZED LIPOSOMES BY TUMOR CELLS.
AU LESERMAN L D; WEINSTEIN J N; BLUMENTHAL R; TERRY W D
CS CENT. IMMUNOL., INST. NATL. SANTE RECH. MED. CENT., NATL. RECH. SCI. MARSEILLE-LUMINY, CASE 906, 13288 MARSEILLE CEDEX 2, FR.
SO PROC NATL ACAD SCI U S A 77 (7). 1980. 4089-4093. CODEN: PNASA6 ISSN: 0027-8424
LA English
AB Specific **receptor**-mediated **delivery** of the contents of small, sonicated liposomes was studied with 3 murine tumor cell types: an Ig[immunoglobulin]G Fc **receptor-negative** nonphagocytic line [lymphoma] (EL4); an Fc **receptor-positive** phagocytic line (P388D1); and an Fc **receptor-positive** nonphagocytic [leukemia] line (P388). The liposomes (formed from phosphatidylcholines, cholesterol and dinitrophenyl-substituted phosphatidylethanolamine) contained carboxyfluorescein as a fluorescent marker and methotrexate as a pharmacologic agent. Binding and internalization of the liposomes were observed by fluorescence microscopy and measured by flow microfluorometry. The hapten-derivatized lipid was used as a binding point on the liposome for the antibody-combining site of the

immunoglobulin. In the presence of IgG anti-dinitrophenyl, but not F(ab')2 or IgA anti-dinitrophenyl, liposomes bound to the Fc **receptor**-bearing cells. The liposomes underwent endocytosis by the P388D1 cells and, to a lesser extent, by the P388 cells. As measured by depression of [3H]deoxyuridine incorporation, methotrexate in IgG-opsonized liposomes had a much greater pharmacologic effect on the P388D1 cells than did the same amount in unopsonized liposomes or in free solution. An appropriately chosen drug, incorporated in liposomes, can exert its effect on a cytoplasmic **target** after endocytosis. P388 **cells** showed a moderate effect of the drug in liposomes. Neither P388 nor P388D1 cells bound or ingested unopsonized liposomes, and the Fc **receptor-negative** EL4 line neither bound nor ingested opsonized liposomes. Specific interaction of opsonized liposomes with the cells' IgG Fc **receptor** were demonstrated. [**Receptor**-mediated endocytosis can result in pharmacologically effective **delivery** of an anti-tumor agent to the cytoplasmic compartment.]

L37 ANSWER 1 OF 7 BIOSIS COPYRIGHT 1996 BIOSIS
AN 96:230304 BIOSIS
DN 98794433
TI Folate-targeted, **anionic** liposome-entrapped polylysine-condensed DNA for tumor cell-specific gene transfer.
AU Lee R J; Huang L
CS Lab. Drug Targeting, Dep. Pharmacol., Univ. Pittsburgh Sch. Med., Pittsburgh, PA 15261, USA
SO Journal of Biological Chemistry 271 (14). 1996. 8481-8487. ISSN: 0021-9258
LA English
AB We have developed a lipidic gene transfer vector, LPD-II, where DNA was first complexed to polylysine at a ratio of 1:0.75 (w/w) and then entrapped into folate-targeted pH-sensitive **anionic** liposomes composed of dioleoyl phosphatidylethanolamine (DOPE)/cholesteryl hemisuccinate/folate-polyethlene glycol-DOPE (6:4:0.01 mol/mol) via charge interaction. LPDII transfection of KB cells, a cell line overexpressing the tumor marker folate receptor, was affected by both the lipid to DNA ratio and the lipid composition. At low lipid to DNA ratios (eg. 4 and 6), LPDII particles were **positively** charged; transfection and **cellular uptake** levels were independent of the folate receptor and did not require a pH-sensitive lipid composition. Meanwhile, transfection and uptake of **negatively** charged LPDII particles, i.e. those with high lipid to DNA ratios (eg. 10 and 12), were folate receptor-dependent and required a pH-sensitive lipid composition. The transfection activity of LPDII was lost when the inverted cone-shaped DOPE was replaced by dioleoyl phosphatidylcholine. LPDII particles with lipid to DNA ratios of 4, 6, 10, and 12 were apprx 20-30 times more active than DNA cntdot 3-beta-(N-(N',N'-dimethylethane)carbamoyl)cholesterol **cationic** liposome complexes in KB cells and were much less cytotoxic. On the sucrose gradient, LPDII particles had a migration rate in between those of the free DNA and the DNA cntdot polylysine complex. An electron micrograph of LPDII showed a structure of spherical particles with a **positively** stained core enclosed in a lipidic envelope with a mean diameter of 74 +- 14 nm. This novel

gene transfer vector may potentially be useful in gene therapy for tumor-specific **delivery**.

L37 ANSWER 2 OF 7 BIOSIS COPYRIGHT 1996 BIOSIS
AN 91:275887 BIOSIS
DN BA92:8502
TI HEPATIC DISPOSITION CHARACTERISTICS OF ELECTRICALLY CHARGED MACROMOLECULES IN RAT IN-VIVO AND IN THE PERFUSED LIVER.
AU NISHIDA K; MIHARA K; TAKINO T; NAKANE S; TAKAKURA Y; HASHIDA M; SEZAKI H
CS FAC. PHARMACEUTICAL SCI., KYOTO UNIV., SAKYO-KU, KYOTO 606, JAPAN.
SO PHARM RES (N Y) 8 (4). 1991. 437-444. CODEN: PHREEB ISSN: 0724-8741
LA English
AB The effect of electric charge on the hepatic disposition of macromolecules was studied in the rat. Charged derivatives of dextran (T-70) and bovine serum albumin (BSA), mitomycin C-dextran conjugates (MMC-D), and lactosaminated BSA (Lac-BSA) were employed as model macromolecules. After intravenous injection, **cationic** macromolecules were rapidly eliminated from plasma because of their extensive hepatic uptake, while **anionic** and neutral macromolecules were slowly eliminated. **Cationic** macromolecules were recovered from parenchymal and nonparenchymal hepatic **cells** at a **cellular uptake** (per unit **cell** number) ratio of 1.4-3.2, while that of Lac-BSA was 14. During liver perfusion using a single-pass constant infusion mode, **cationic** macromolecules were continuously extracted by the liver, with extraction ratios at steady-state (Ess) ranging between 0.03 and 0.54, whereas **anionic** and neutral macromolecules were almost completely recovered in the outflow at steady state. The Ess for **cationized** BSA (Cat-BSA) and **cationic** MMC-Dcat were concentration dependent and decreased at low temperatures and in the presence of colchicine and cytochalasin B. The possible participation of the internalization process in the uptake of **cationic** macromolecules by heptocytes was suggested.

L37 ANSWER 3 OF 7 BIOSIS COPYRIGHT 1996 BIOSIS
AN 90:176425 BIOSIS
DN BA89:93595
TI TRANSFER OF PREFORMED TERMINAL C5B-9 COMPLEMENT COMPLEXES INTO THE OUTER MEMBRANE OF VIABLE GRAM-**NEGATIVE** BACTERIA EFFECT ON VIABILITY AND INTEGRITY.
AU TOMLINSON S; TAYLOR P W; LUZIO J P
CS ADDENBROOKE'S HOSP., HILLS RD., CAMBRIDGE CB2 2QR, UK.
SO BIOCHEMISTRY 29 (7). 1990. 1852-1860. CODEN: BICHAW ISSN: 0006-2960
LA English
AB An efficient fusion system between Gram-**negative** bacteria and liposomes incorporating detergent-extracted C5b-9 complexes has been developed that allows **delivery** of preformed terminal complexes to the cell envelope (Tomlinson et al., 1989b). Fusion of Salmonella minnesota Re595 and Escherichia coli 17 with C5b-9-incorporated liposomes resulted in the transfer of 1900 C5b-9 complexes to each target bacterial cell. No loss in viability of bacteria was observed following fusion, even though the deposition of 900 complexes onto the envelope following exposure to lysozyme-free serum effected a greater than 99% loss of viability. Increased sensitivity to antibiotics normally excluded from the cell by an integral outer membrane (OM), as well as the ability of the

chromogenic substrate PADAC to gain access to periplasmically located .beta.-lactamase, indicated that transferred C5b-9 complexes functioned as water-filled channels through the OM. A similar conclusion was drawn from measurements demonstrating the **uptake** by **cells** of the lipophilic **cation** tetraphenylphosphonium (bromide), a result further indicating that the membrane potential across the cytoplasmic membrane was maintained following C5b-9 transfer to the OM. Examination of S. minnesota Re595 by electron microscopy revealed no obvious difference between cells exposed to lethal concentrations of lysozyme-free serum and cells following fusion with C5b-9-incorporated liposomes. These data suggest either that there are critical sites in the OM to which liposome-**delivered** C5b-9 complexes are unable to gain access or that bacterial cell death is related to events occurring during polymerization of C9 on the cell surface.

L37 ANSWER 4 OF 7 BIOSIS COPYRIGHT 1996 BIOSIS
AN 88:180447 BIOSIS
DN BA85:92549
TI ENHANCED ANTIPROLIFERATIVE ACTION OF INTERFERON TARGETED BY BISPECIFIC MONOCLONAL ANTIBODIES.
AU ALKAN S S; TOWBIN H; HOCHPEPPEL H-K
CS PHARMACEUTICALS RES. DIV., INFLAMMATION/ALLERGY, ROSENTAL-1056.4.07, CH-4002 BASEL, SWITZERLAND.
SO J INTERFERON RES 8 (1). 1988. 25-33. CODEN: JIREDJ ISSN: 0197-8357
LA English
AB It has previously been shown that interferon (IFN) can be coupled covalently to tumor-specific monoclonal antibodies (mAb) and that the in vitro antiviral and antiproliferative action of these IFN-mAb conjugates is superior to that of uncoupled IFN. We now report a different mode of IFN **delivery**, i.e., via bispecific mAbs, avoiding chemical coupling of IFN. Bispecific mAbs were prepared by cross-linking two mAbs with SPDP, mAb1 being specific for an idiotype of a hybridoma cell-surface immunoglobulin and mAb2 specific for an IFN. Alternatively, Fab' fractions of mAb1 and mAb2 were coupled by disulfide formation to produce F(ab')2. Binding capacity and specificity of both arms of the mAb conjugates were first demonstrated by a solid-phase radioimmunoassay using idiotype-**positive** mAb as test antigen and 125I-labeled hybrid IFN-.alpha.B/D. Secondly, hybridomas either idiotype **positive** or **negative** were incubated with bispecific mAbs (mAb1-mAb2 or Fab'1-Fab'2) and 125I-labeled IFN at 4.degree. C. After washing away unbound reagents, the **uptake** of radioactivity into **cells** was determined. Additionally, the antiproliferative action of cold or labeled IFN targeted via different modes was assessed by an [3H]TdR incorporation method. Results showed that bispecific mAbs could specifically **deliver** IFN to the target cells and also inhibit their growth in vitro. Furthermore, targeting IFN by any of the three methods, IFN-mAb, mAb1-mAb2, or Fab'1-Fab'2 enhanced its in vitro antiproliferative potency compared to IFN alone.

L37 ANSWER 5 OF 7 BIOSIS COPYRIGHT 1996 BIOSIS
AN 88:155758 BIOSIS
DN BA85:79411
TI A NOVEL IN-VIVO FOLLICULAR DENDRITIC CELL-DEPENDENT ICCOSOME-MEDIATED MECHANISM FOR **DELIVERY** OF ANTIGEN TO ANTIGEN-PROCESSING CELLS.

AU SZAKAL A K; KOSCO M H; TEW J G
CS DEP. ANAT., DIV. IMMUNOL., P.O. BOX 709, MCV STATION, MCV/VCU, RICHMOND, VA. 23298.
SO J IMMUNOL 140 (2). 1988. 341-353. CODEN: JOIMA3 ISSN: 0022-1767
LA English
AB Recent scanning electron microscopic studies on isolated follicular dendritic cells (FDC) showed that dendrites of certain FDC were "beaded" i.e., consisting of a series of interconnected immune complex coated bodies (termed "iccosomes," measuring 0.3 to 0.7 .mu.m diameter). In vitro these iccosomes detach from one another with ease. The major objectives herein were to establish whether these structures can be detected in sections and whether iccosomes serve to disseminate antigen in vivo. Beginning at day 1, the time point used for isolating beaded FDC, the popliteal lymph nodes of immune C3H mice were studied with light and transmission electron microscopy for 2 wk (i.e., at days 1, 3, 5, 8, and 14) after hind footpad injection of the histochemically detectable antigen, horseradish peroxidase (HRP). Iccosomes (0.25 to 0.38 .mu.m diameter), contoured by a peroxidase (PO)-**positive** coat of HRP-anti-HRP complexes, were first detected by transmission electron microscopy at day 1 adjacent to cell bodies of certain FDC. Within their limiting membrane they contained flocculent material that was PO **positive**. At day 3 by light microscopy, germinal centers were seen enlarged and the antigen-retaining reticulum, composed of antigen-bearing FDC, appeared diffuse. This coincided with the transmission electron microscopic visualization of a dispersed state of iccosomes among the follicular lymphocytes. At that time iccosomes were seen attached to the surface of lymphocytes via PO-**positive** immune complexes and were surrounded by microvillous processes of these cells. Germinal center lymphocytes and tingible body macrophages both responded to contact with iccosomes by endocytosis. Antigen-containing tingible body macrophage were most conspicuous by light microscopy at day 5, when transmission electron microscopy showed that the majority of germinal center lymphocytes contained endocytosed HRP in secondary lysosome-like granules associated with the Golgi apparatus. The number of dispersed iccocomes was markedly reduced by day 5. In contrast injected with HSA, a PO-**negative** antigen, lymphocytes and tingible body macrophages were PO-**negative**. The presence of antigen in both cell types was confirmed through the use of a gold-conjugated antigen (goat IgG). simultaneous immunoperoxidase labeling of the same tissues with anti-Ia showed the gold conjugate containing B cells to be Ia+. Antigen-**positive** B cells and tingible body macrophages were greatly reduced in numbers by day 14, suggesting the intracellular fragmentation of the antigen. These observations and the tightening of the antigen-retaining convolutions of filiform (non-beaded) dendrites of FDC created the overall impression that by day 14 the germinal center response was winding down. This study demonstrated a novel FDC-dependent iccosome-mediated mechanism for **delivering** antigen to germinal center B cells and tingible body marcophages, which should be capable of processing and presenting antigen to T **cells**. Observations of antigen **uptake** suggested an apparent lysosomal fragmentation of antigen for possible re-expression via Golgi functions. Germinal center B cells and tingible body macrophages are likely candidates for antigen processing and presentation to germinal center T helper cells in vivo. This process may help explain the development of immunologic memory and the high antibody titers associated with the

secondary immune response.

L37 ANSWER 6 OF 7 BIOSIS COPYRIGHT 1996 BIOSIS
AN 86:437617 BIOSIS
DN BA82:103805
TI IN-VITRO GENOTOXICITY STUDIES USING COMPLEX HYDROPHOBIC MIXTURES EFFICIENT **DELIVERY** OF A PETROLEUM SAMPLE TO CULTURED C-3H-10T-1-2 CELLS VIA LIPID VESICLE INCORPORATION.
AU VON HOFE E H; BILLINGS P C; HEIDELBERGER C; LANDOLPH J R
CS NORRIS CANCER HOSP. AND RES. INST., COMPREHENSIVE CANCER CENTER, UNIV. SOUTHERN CALIF. SCH. MED., 2025 ZONAL AVE., LOS ANGELES, CALIF. 90033.
SO ENVIRON MUTAGEN 8 (4). 1986. 589-610. CODEN: ENMUDM ISSN: 0192-2521
LA English
AB Petroleum fractions are a diverse group of extremely hydrophobic mixtures, some of which display strong carcinogenicity in animal skin painting experiments. Interpretation of in vitro genotoxicity experiments with these samples is complicated by inefficient **delivery** of these hydrophobic substances inside target cells. We therefore developed methods to assess and improve the efficiency of **delivering** a petroleum sample (Matrix, A.P.I. 81-17) to cultured C3H/10T1/2 cells for genotoxicity studies via lipid vesicle incorporation. Three radiolabeled compounds (14C-benzo(a)pyrene, 14C-decane, and 14C-naphthalene) of widely differing volatilities, broadly representative of the spectrum of compounds in petroleum samples, were separately added to Matrix. Lipid vesicles containing Matrix and radiolabeled compounds were prepared by the classical methods for preparing neutral, **positive**, and **negatively** charged multilamellar and unilamellar liposomes. Of these, the classical methods for preparing neutral unilamellar liposomes were the most successful for **delivering** radiolabeled compounds in Matrix to cells. Vesicles optimal for the **delivery** of tracers in Matrix were prepared with DSPC:cholesterol:lyso-PC (8.8:0.8:0.4, molar ratio) in a Matrix to lipid ratio of 31:69 (w/w). This new method of **delivery** resulted in proportional, dose-dependent, and reproducible **uptake** of all tracers. Further, **cells** treated with this preparation took up 2.5-fold more 14C-decane, 1.5-fold more 14C-BaP, and 18-fold more 14C-naphthalene added to Matrix than did cells treated with Matrix emulsified in tissue culture medium. In contrast, tracers were not taken up in a proportional or reproducible manner when emulsions were used, and in fact, uptake of 14C-naphthalene was consistently very small. Two petroleum fractions, C2029188 and C3029194, were 4- and 6-fold more cytotoxic, respectively, when **delivered** to C3H/10T1/2 cells by lipid vesicles than emulsions. The carcinogenic petroleum fraction C50292202 induces type II transformed foci in C3H/10T1/2 cells when cells were treated with C50292202 incorporated into lipid vesicles. The methods for lipid vesicle incorporation described here are effective in **delivering** hydrophobic petroleum fractions to cells and provide an alternative to the current inefficient and artifactual methods of emulsification currently used. With further validation and standardization, lipid vesicle incorporation of petroleum fractions and treatment of cells with these vesicles should be useful for studying the genotoxicity of complex hydrophobic mixtures in cell culture systems.

L37 ANSWER 7 OF 7 BIOSIS COPYRIGHT 1996 BIOSIS

AN 83:219967 BIOSIS
DN BA75:69967
TI UPTAKE OF LIPOSOMES AND LIPOSOME ENCAPSULATED MURAMYL DI PEPTIDE BY HUMAN PERIPHERAL BLOOD MONOCYTES.
AU MEHTA K; LOPEZ-BERESTEIN G; HERSH E M; JULIANO R J
CS DEP. OF PHARMACOL., THE UNIV. OF TEX. MED. SCH. AT HOUSTON, P.O. BOX 20708, HOUSTON, TX 77205.
SO J RETICULOENDOTHEL SOC 32 (2). 1982. 155-164. CODEN: JRSODF ISSN: 0033-6890
LA English
AB The interaction of multilamellar liposomes with human peripheral blood monocytes, cultured in vitro, were examined. Phagocytic engulfment is the principal mechanism by which the liposomes are taken up by these cells. Studies with radiolabeled liposomes revealed that they are taken up by monocytes as intact structures. The uptake is temperature sensitive and is affected by inhibitors of glycolysis and of microfilament activity. Monocytes take up **negatively** charged vesicles more rapidly than **positively** charged (3-fold) vesicles or neutral vesicles (5-fold). Increase in **negative** charge of liposomes enhances their **uptake** by the **cells**, but increased saturation of the phospholipids results in decreased uptake. Liposomes provide an effective means of enhancing the uptake of MDP [muramyl-L-alanyl-D-isoglutamine] derivatives (3H-norMDP) by monocytes, with a 20-fold greater uptake of liposome encapsulated drug than of the free compound. Monocytes do not degrade the 3H-norMDP that they have internalized and radiolabel is only slowly released from the cells. These observations suggest a pharmacokinetic basis for the use of lipid vesicles as a system for the **delivery** of immunomodulating drugs to monocytes.

=> D HIS

(FILE 'HCAPLUS' ENTERED AT 07:20:14 ON 25 SEP 96)
DEL HIS Y
L1 48268 S DELIVER?
L2 622 S L1 AND (POS OR POSITIV? OR POLYCATION? OR CATION?) AND
L3 17 S L2 AND (POLYMAINE# OR SPERMIDINE? OR POLYLYSINE OR POLY
L4 52 S L2 AND (POLYNUCLEO? OR DNA OR NUCLEIC OR OLIGONUCLEO?)
L5 5 S L2 AND POLYAMINE?
L6 19 S L3 OR L5
L7 9 S L6 AND L4
L8 21 S L2 AND CELL####(3A)TARGET?
L9 6 S L8 AND POLYMER?
L10 13 S L7 OR L9
L11 7 S L8 AND (L3 OR L4 OR L5)
L12 13 S L7 OR L9 OR L11
L13 7 S L4 AND CELL####(3A)TARGET?
L14 1 S L13 AND (CONTRAST? OR IMAG?)
L15 855 S L1 AND CELL####(3A)TARGET?
L16 263 S RECEPTOR? AND L15
L17 12 S L16 AND L2
L18 8 S L2 AND CELL####(4A)UPTAKE?
L19 0 S L18 AND HYDROPHOB?
L20 0 S L14 NOT L12
L21 0 S L13 NOT L12
L22 8 S L17 NOT L12
L23 6 S L18 NOT (L12 OR L17)
SAV JONES/L ALL

FILE 'WPIDS' ENTERED AT 08:24:28 ON 25 SEP 96
L24 8 S L12
L25 1 S L14
L26 1 S L17
L27 6 S L18
L28 10 S L24-L27

FILE 'MEDLINE' ENTERED AT 08:31:41 ON 25 SEP 96
L29 9 S L12
L30 0 S L14
L31 9 S L17
L32 6 S L18
L33 21 S L29-L32

FILE 'BIOSIS' ENTERED AT 08:37:37 ON 25 SEP 96
L34 11 S L12
L35 0 S L14
L36 10 S L17
L37 7 S L18
L38 23 S L34-L37

FILE 'HCAPLUS' ENTERED AT 08:42:45 ON 25 SEP 96
L39 27 S L12 OR L14 OR L17 OR L18

FILE 'BIOSIS' ENTERED AT 08:42:53 ON 25 SEP 96